ABSTRACT

GROWE, ANTHONY MICAJAH. Effects of Simulated Dicamba Drift on Maturity Group V and VI Soybean Growth and Yield. (Under the direction of Wesley Everman).

Dicamba-tolerant crop varieties have the potential to become utilized in North Carolina as a tool to control glyphosate-resistant weeds. Because of North Carolina's diverse agricultural landscape, there is growing concern over the potential for off-site deposition of dicamba to sensitive and valuable crops. A literature review of previous research has determined that soybean, commonly glyphosate- or glufosinate-tolerant varieties, are highly sensitive to dicamba (Johnson et al. 2012, Griffin et al. 2013, Wax et al. 1969). Tank contamination, spray drift, and volatilization of dicamba can cause injury and reduce soybean yield. To date, there has been little information reported on soybean varietal responses to sub-lethal doses of dicamba. The objective of this study was to evaluate the effects of sub-lethal rates of dicamba on MG V and MG VI (maturity groups V and VI) soybean cultivars at the vegetative and reproductive growth stages. These maturity groups are commonly grown in North Carolina and varieties were selected from the North Carolina Official Variety Testing list. Separate experiments were conducted with the two maturity groups. Effects of dicamba were determined by collecting visual injury ratings, height reductions, and yield. Four varieties within each maturity group were subjected to a range of sub-lethal dicamba rates (0.2 to 12.5% of the recommended rate for dicamba-tolerant soybean) during the V4 (vegetative growth stage with three completely unrolled trifoliate leaves) and R2 (reproductive stage at full flower) growth stages. Each experiment was conducted in 2015 and 2016 on a Portsmouth loam soil in Kinston, NC and a Rains sandy loam soil in Rocky Mount and Lewiston, NC.

Increasing height reduction and visual injury were associated with increasing

dicamba rates. Height reductions 14 and 28 DAT were generally greater from dicamba

applied at the vegetative growth stage compared to the reproductive stage regardless of year

and soil type. Variability in yield reductions indicated a strong environmental influence on

soybean response to sub-lethal rates of dicamba. For both experiments, soil type impacted

soybean yield response to dicamba. On sandy loam soils, a greater soybean yield loss was

observed with dicamba applied the R2 stage compared with application to the V4 stage. On

the loam soil, yield reduction was either greater when dicamba was applied at the V4 stage or

there was no significant timing effect. Depending on year, soil type, and growth stage, a

significant varietal response was recorded for each experiment. For the MG V experiment,

AG5935 sustained greater yield reduction than S56RY84 or P 5610 RY when dicamba was

applied at the R2 stage in 2016. For the MG VI experiment, SS 6810N R2 demonstrated the

greatest sensitivity when dicamba was applied at the R2 stage in 2016. In some cases, a

varietal response was absent.

While data indicated injury ratings and height reductions were moderately correlated

with yield loss, these measurements are not 100% reliable in predicting yield loss after a drift

event. Not only do the data suggest there is a varietal response, there are also implications

that environmental factors (such as soil type, precipitation, temperature) heavily influence

soybean response to dicamba drift. Factors such as humidity, application time of day,

rainfall, and ambient and soil temperature were analyzed but revealed no patterns that can

explain the variability of the results.

With such variability between environments, yield loss may be difficult to accurately

predict. With the recent release of dicamba-tolerant soybean cultivars and North Carolina's

diverse agricultural landscape, injury to highly sensitive and valuable crops planted in close proximity can be expected. When using dicamba as a weed management option, it is important that growers abide by both federal and state labels and use the best judgement possible when making applications to minimize off-target deposition. Using appropriate nozzles, boom heights, travel speeds, and registered formulations plus monitoring wind speed and direction are practices applicators should follow to minimize off-target deposition of dicamba.

Effects of Sub-lethal Rates of Dicamba on Maturity Group V and VI Soybean Growth and
Yield.

by
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A thesis submitted to the Graduate Faculty of
North Carolina State University
in partial fulfillment of the
requirements for the degree of
Master of Science

Crop and Soil Science

Raleigh, North Carolina

2017

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DEDICATION

This thesis is dedicated to my parents, David and Donna Growe. They have showed me that success is achieved through hard work and diligence. They have always encouraged me to chase my dreams no matter how big they are. Words cannot describe how thankful I am for everything they have done as parents. I also dedicate this thesis to my wife and best friend, Shelby Growe. Without their continuous love, support and encouragement, this achievement would not be possible.

BIOGRAPHY

Anthony Growe is a master's student at NC State in the Department of Crop and Soil Science under the direction of Dr. Wesley Everman. He was born and raised near Hamlet, NC and always had the dream to attend NC State. Helping his father with their small cattle operation, Anthony understood the importance of North Carolina agriculture. While working as an hourly undergraduate student for Dr. Everman, Anthony developed a passion for weed science in field crops. To date, Anthony has assisted with the completion of countless weed control efficacy trials and extension demonstrations. He also received the Becky and Neil Rhodes Weed Science Graduate Education Award, delivered several presentations at weed science conferences, placed second in the Northeast Weed Science Society weed team competition, and received the Weed Science Society of North Carolina Outstanding Graduate Student Award. After graduation, Anthony will work as an extension agent in Cumberland County. His goals are to connect growers with research derived information through education and increase sustainability and profitability of agriculture. In his spare time, Anthony enjoys working in the yard, hunting, fishing, and spending quality time with his wife, family and friends.

ACKNOWLEDGMENTS

I would like to thank Dr. Everman for giving me the opportunity to further my knowledge and education while pursuing this degree. I am grateful to be given a chance to take on a new yet unfamiliar area of study in crop science. I would also like to thank my committee members, Dr. Dunphy and Dr. York, for their direction and advice. I also would like to thank the research team who helped in the completion of this research: Liam Vincent, Adam Blackmore, John Sanders, Mitch Schroder, Drake Copeland, Brandon Schrage, Patrick Whitt, Manish Bansal, Theresa Reinhardt, Zack Taylor, Alex Knight, and Thierry Besancon. They all earned a piece of this achievement through their hard work and dedication required to complete this research. Additionally, this research would not have been successful without the outstanding help from the staff of the research stations where we worked; specifically, the Upper Coastal Plain Research Station in Rocky Mount, NC; the Peanut Belt Research Station in Lewiston-Woodville, NC; and the Caswell Research Farm in Kinston, NC. I am beyond grateful for those that have helped me on this journey.

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Literature Review

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Soybean *(Glycine max L.)* Production in the United States and North Carolina

In 2014, the United States produced over one hundred million metric tons of soybean and was the number one producer in the world (FAO 2017). According to the United States Department of Agriculture, soybean is one of the most dominant sources of animal feed. It accounts for 90% of oilseed production and is the second most widely produced crop in the United States (USDA-NASS 2016). A record-breaking 33 million hectares of soybean were harvested nationwide in 2016 (USDA-NASS 2016).

Like other grains, the majority of soybean produced in North Carolina is processed for oil and used to feed livestock (North Carolina soybean Producers Association 2017). In 2012, the state ranked first in poultry and egg sales and second in pig sales (USDA-NASS 2016). To fuel the livestock production, soybean meal has been a desirable protein source due to its relatively high protein content of 44 to 50 %, its availability, and consistently competitive price (Brookes 2000). In 2012, North Carolina ranked fifteenth in the nation for soybean hectares planted (USDA-NASS 2016). In 2015, 671,800 soybean hectares were harvested and ranked second in the total production sales of crop commodities in North Carolina with an estimated monetary value of 486 million dollars (USDA-NASS 2016). The introduction of herbicide tolerance traits, narrow-row planting, and crop rotation practices have allowed for yield improvements which favored expansion of soybean acreage (USDA-NASS 2016). The statistics presented above illustrate the importance of soybean production to the livestock market and the state's economy as a whole.

In 1996, Monsanto introduced Roundup Ready® soybean which is a transgenic crop tolerant of the non-selective herbicide, glyphosate. Roundup Ready® cotton was released in 1997 followed by corn (*Zea mays* L.) in 1998 (Johnson et al. 2012). With glyphosate's low toxicity and ease of handling and mixing, glyphosate-tolerant crops offered many advantages in controlling weeds compared to conventional systems and was adopted worldwide. Glyphosate's nonselective nature allowed growers to manage a wide spectrum of weeds with a few applications of the product each season. The replacement of traditional weed control practices and chemistries, such as residual herbicides, with programs consisting only of glyphosate led to selection for glyphosate-resistant weed biotypes (Culpepper et al. 2006). In 2005, glyphosate-resistant Palmer amaranth [*Amaranthus palmeri* S. Wats.] was confirmed in Georgia followed by North Carolina (Culpepper et al. 2008). Today, glyphosate-resistant Palmer amaranth has been confirmed in 26 states (Heap 2017). Other species with glyphosate-resistant biotypes in the United States include horseweed [*Conyza canadensis* (L.) Cronquist], tall waterhemp [*Amaranthus tuberculatus* (Moq.) Sauer], Italian ryegrass [*Lolium perenne* spp. m*ultiflorum* (Lam.) Husnot], common ragweed (*Ambrosia artemiisifolia* L.), giant ragweed (*Ambrosia trifida* L.), goosegrass [*Eleusine indica* (L.) Gaertn.], johnsongrass [*Sorghum halepense* (L.) Pers.], and kochia [*Kochia scoparia* (L.) Schrad.] (Heap 2017).

Dicamba and its Impact on Soybean Production

Dicamba (3, 6-dichloro-2-methoxybenzoic acid) is an herbicide in the benzoic acid chemical family and in the acid form, has a vapor pressure of 4.5 mPa at 25°C. It is characterized as a moderately volatile compound (Bunch and Gervais 2012) and was discovered by S. B. Richter in 1958 but the mechanism of action is not fully understood. It is believed to be similar to the endogenous auxin indoleacetic acid (IAA), which is a naturally occurring hormone in plants (Kelley and Riechers 2007). While the concentration of this endogenous auxin and its effects are regulated under most conditions, auxinic herbicides interrupt the natural regulatory processes of sensitive plants to cause an uncontrolled auxin response (Kelley and Riechers 2007). Auxin is first perceived by the plant through its transport inhibitor response (TIR1) proteins. These proteins are required for the degradation of Aux/IAA transcriptional repressors (Gleason et al. 2011). In plant physiology, it is understood that high auxin levels induce the biosynthesis of abscisic acid (ABA). ABA induces stomatal closure, which causes carbon fixation by photosynthesis to cease (Cobb and Reade 2010). ABA combined in the presence of light is also thought to cause accumulation of hydrogen peroxide resulting in oxidative damage which contributes to phytotoxicity (Cobb and Reade 2010).

Dicamba may be applied burndown, preemergence (PRE), and postemergence (POST) in corn and small grains to control broadleaf weeds at rates of 280 to 560 g ae ha^{-1} (depending on soil type) and is also used in fallow land, pastures, and turf. In 2015, Monsanto released Roundup Ready 2 Xtend® soybean and Bollgard II® XtendFlex® cotton (*Gossypium*

hirsutum L.) varieties. This technology will allow the use of labeled dicamba products and glyphosate POST in soybean and cotton. Monsanto's and DuPont's new dicamba products, Xtendimax™ (Monsanto Company, St. Louis, MO 63167) and FeXapan™ (Dupont, Wilmington, DE 19898) contain the diglycolamine (DGA) salt of dicamba plus the proprietary ingredient VaporGrip Technology® which is intended to reduce volatility. Additionally, BASF's new dicamba product, Engenia® (BASF Corporation, Research Triangle Park, NC 27709) contains a N-Bis-(aminopropyl) methylamine (BAPMA) salt which is heavier in molecular weight to reduce vapor drift. With concerns over the off-target movement of dicamba, applying older, more volatile formulations, such as Clarity® (BASF Corporation, Research Triangle Park, NC 27709) or Banvel® (Arysta LifeScience North America LLC, Cary, NC 27513) is not permitted. Clarity contains the DGA salt of dicamba and Banvel contains the dimethylamine (DMA) salt of dicamba. Egan and Mortensen (2012) reported reduced vapor drift with DGA versus DMA formulations. Although the DGA salt is less volatile than the DMA, both of these products can only be applied as a burndown before planting soybean; any POST applications to dicamba-tolerant soybean or cotton are considered off-label. Unfortunately, these new dicamba products did not receive EPA (Environmental Protection Agency) approval when the dicamba-tolerant varieties were released for commercial use. In attempts to control infestations of herbicide-resistant weeds, there were reports of growers planting dicamba-tolerant cultivars and making illegal applications of old formulations such as Clarity® before new formulations of dicamba were approved. Because of the moderately volatile nature of dicamba and its unique symptomology, there has been an increase of dicamba drift complaints in the mid-South

states. "More than 100 farmers in Missouri have filed formal complaints with the state's Department of Agriculture and in Arkansas, 25 complaints have been filed" (Bennett 2016).

Dicot crops such as soybean, cotton (*Gossypium hirsutum* L.), peanut (*Arachis hypogaea* L.), tobacco (*Nicotiana tabacum* L.), and most vegetable crops are very sensitive to low rates of dicamba (Al-Khatib and Peterson 1999; Batts 2015; Dittmar et al. 2016; Johnson et al. 2012). If growers adopt the use of dicamba on dicamba-tolerant crops, non-tolerant soybean varieties in adjacent fields are at risk for injury and possible yield loss. Previous research has demonstrated that yields of non-tolerant soybean can be impacted by minute amounts of dicamba that can move off-target through spray drift or volatility. Brehens and Lueschen (1979) observed dicamba injury, caused by vapor drift, to potted soybean plants placed 60 m from a corn field 1 hour after it was treated with dicamba. Large portions of soybean acres hectares can be exposed to dicamba through volatility (vapor drift), spray drift, and sprayer contamination. With numerous factors such as herbicide rate, wind speed, travel speed, boom height, and nozzle type, calculating a "drift or volatility rate" can be difficult. Downwind drift deposits from an unshielded ground rig sprayer have been reported to range from 1 to 8% depending on nozzle type and wind speed (Bode 1988, Maybank et al. 1978). Boerboom (2004) detected 0.63% of a 560 g ha^{-1} dicamba use rate from the spray boom after an ammonia cleaning solution had been used. Cundiff et al. (2017) reported that hose material impacts the amount of dicamba residues that remain in the sprayer. Data showed that synthetic rubber hoses retained 0.5% of a 1X rate of dicamba compared to polyethylene blended hoses which retained 0.06%. Egan and Mortensen (2012) quantified dicamba vapor at an average concentration of 0.56 g ha^{-1} 21 m away from the

treated plot. Weidenhamer et al. (1989) observed a 10% yield loss when 0.4 g ha^{-1} of dicamba was applied to mid-bloom soybean. Growers should use caution and exercise best management practices, such as slower travel speed, coarse droplet size, and lower boom height, when using auxin herbicides for a weed control option.

Dicamba symptomology on soybean is typical of auxin herbicides. Soybean exhibits severe shoot and petiole epinasty and leaf cupping, stunting, curling, strapping, and distortion (Al-Khatib and Peterson 1999). Dicamba's effects on soybean have been well documented over the last 50 years. Wax et al. (1969) reported that the injury to soybean from dicamba exceeded that from equivalent rates of 2, 4-D. Attempts have been made to develop injury rating and height reduction scales to predict yield loss. Al-Khatib and Peterson (1999) reported over 50% injury to soybean plants 14 d after a 17 g ha^{-1} rate of dicamba was applied at V2 to V3 growth stage. When V2 to V3 soybean was exposed to one-half the labeled rate of dicamba (280 g ha^{-1}), a height reduction of 72% was observed (Griffin et al. 2013). After reviewing research that utilized these methods to predict yield loss, there is some disagreement on the accuracy of the indicator scales. Auch and Arnold (1978) concluded that one could not always expect yield reduction when plant height is reduced from dicamba injury. In contrast, Weidenhamer et al. (1989) reported that "height reductions and plant morphology are good predictors of yield reductions from dicamba injury with the exception of Elf [a determinate soybean variety] treated at the mid-bloom stages." Due to determinate soybean's growth habit, height reductions may not reflect yield loss as accurately as indeterminate varieties at the reproductive stages. Johnson et al. (2012) treated soybean with 1/2, 1/8, 1/32, 1/128, and 1/512 of the normal dicamba rate of 560 g ha^{-1}. Results showed

that injury ratings associated with soybean yield loss ranged from 30 to 100% 2 weeks after

treatment (WAT). Johnson et al. (2012) concluded that the coefficients for dicamba were

greater than or equal to 0.6, indicating that dicamba injury symptomology was a moderate

predictor of yield. In one experiment, Robinson et al. (2013) assessed dicamba injury 28

days after treatment (DAT) and the regression analyses of yield loss to injury ratings 14 and

28 DAT revealed a 0.91 R^2 value (Robinson et al. 2013). The authors inferred that using

visual injury scales is a reliable approach to predicting yield loss.

Although height reduction and injury may be moderate indicators of yield loss, growth

stage at the time of dicamba exposure is a factor that impacts soybean's yield response

(Griffin et al. 2013). Research revealed that a 187 g ha^{-1} (1/3 of the labeled 560 g ha^{-1} use

rate in corn) rate of dicamba, applied to V2 to V3 growth stage soybean, reduced yield at

least 75% (Al-Khatib and Peterson 1999). Wax et al. (1969) reported yield reduction of 23%

when 4.4 g ha^{-1} was applied at the mid bloom while 35 g ha^{-1} was required to reduce yield by

20% for the pre-bloom (vegetative) stage. Griffin et al. (2013) indicated a 15 and 36%

reduction in yield when 17.5 g ha^{-1} (1/32 of use rate) of dicamba was applied V4 and R1,

respectively, and concluded that soybean exposed to dicamba during R1 is 2.5 times more

sensitive compared to vegetative exposure. Auch and Arnold (1978) found that an 11 g ha^{-1}

rate reduced yields when applied at the early bloom timing but not vegetative timings.

Similarly, dicamba applied to V3 and R2 soybeans at 0.028, 0.28 and 28 g ha^{-1} reduced yields

by 2 to 67% for the R2 timing, but there was no observed yield loss for V3 applications

(Solomon and Bradley 2014). Robinson et al. (2013) reported that soybean yield loss was

lower when dicamba was applied at vegetative stages and could be attributed to plants having

more time to overcome injuries before reproduction began. In contrast to other literature, Weidenhamer et al. (1989) reported that the differences in yield reductions caused by pre-bloom and mid-bloom applications were smaller than previous studies and attributed it to the later application of the pre-bloom treatments. Also, Kelley et al. (2005) reported 7% yield reduction from dicamba at 0.56 g ha^{-1} applied to V3 stage soybean but no yield reduction with the same rate of dicamba applied to V7 or R2 stage soybean. Dicamba at 5.6 g ha^{-1} reduced yield 6, 12, and 7% when applied to V3, V7, and R2 stage soybeans, respectively. Although previous studies have demonstrated that growth stage does affect soybean's response to dicamba drift, there are some conflicting conclusions on which growth stage is more sensitive to dicamba. More research should be carried out to understand these effects.

In addition to growth stage at time of exposure, other factors also seem to impact soybean injury from dicamba. Greater yield reductions have been reported in drier growing seasons (Andersen et al. 2004; Auch and Arnold 1978; Robinson et al. 2013; Weidenhamer et al. 1989). Al-Khatib and Peterson (1999) observed increased soybean sensitivity to dicamba with high temperatures at time of exposure. Also, certain tank mixes have synergistic effects on both weeds and crops. Kelley et al. (2005) reported synergistic interactions between dicamba at 5.6 g ha^{-1} and normal use rates of glyphosate, imazethapyr, imazamox, and fomesafen. Thus, the impact of dicamba residue in spray equipment could be enhanced when the equipment is used to apply herbicides to non-dicamba-tolerant soybean.

Smith and Caviness (1973) treated 10 soybean varieties with two rates of propanil, a common herbicide used in rice (*Oryza sativa* L.), and found that some varieties were more tolerant than others. 'Davis', 'Hood' and 'York' soybean was damaged more than 'Hill',

'Lee', 'Lee 68', 'Pickett', 'Semmes', 'Bragg' and 'Dare' when damage was measured by reduction in seed yield and by leaf injury (Smith and Caviness 1973). Similar to Smith and Caviness (1973), Wax et al. (1976) evaluated soybean variety response to metribuzin. The authors concluded that there was a differential response of soybean cultivars to metribuzin. Varieties such as 'Atlona', 'Hurrelbrink', 'Norman', 'Portage', 'Semmes', 'Tracy', and 'Vansoy' were extremely sensitive and were not recommended when using metribuzin as a weed control option (Wax et. al 1976).

If soybean has a differential response to photosystem II-inhibiting herbicides, such as propanil and metribuzin there could be one for dicamba as well. Weidenhamer et al. (1989) compared the response of Elf, a determinate variety, and Williams, an indeterminate variety, to dicamba applied at the pre-bloom and mid-bloom stages. Height reduction was greater with Williams than Elf with dicamba applied at the mid-bloom stage. Greater yield reduction also occurred with Williams than with Elf, especially with application at the mid-bloom stage. A response to dicamba may depend on an interaction between growth habit and growth stage at time of exposure. After reviewing the literature, the last variety trial conducted was in 1978 by Auch and Arnold (1978). The objective of the study was to test common soybean varieties grown in South Dakota for dicamba tolerance. Five soybean cultivars, Corsoy, 'Harcor', 'SRF-200', 'Wells' and 'Amsoy 71', were treated with 28 g ha^{-1} at the early bloom, mid-bloom and early pod stages. Results demonstrated that "yield reduction cannot be prevented by the preferential planting of these varieties" (Auch and Arnold 1978). Although the varieties were not tolerant to dicamba, there were varietal responses with statistical significance. When applications were made at the early bloom

stage, yield was reduced in all varieties with the exception of Wells. During the mid-bloom stage, significant yield reduction was not observed for Corsoy and Harcor. These cultivars' response to dicamba applied at different growth stages indicates a varietal influence on soybean response to sub-lethal rates of dicamba. With advances in breeding and biotechnology, there is a possibility of modern soybean cultivars being more susceptible or tolerant to dicamba at various rates and growth stages.

With the use of dicamba in Roundup Ready 2 Xtend® soybean varieties, there is concern of off-target movement to sensitive crops. Research must be done to address the varietal response of soybean to dicamba at vegetative and reproductive stages. Information gained from this research can help growers, extension specialists and agents, and industry representatives understand the effects of dicamba on soybean, a major crop for the seed oil and animal feed industry in North Carolina. Testing the relationship of soybean height reduction to yield loss will determine if it is a sufficient indicator of yield loss. If these techniques are reliable, they can serve as templates for growers and extension agents to predict the severity of yield reduction from dicamba drift.

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Chapter II

Effect of Simulated Dicamba Drift on Maturity Group V Soybean Growth and Yield

Anthony M. Growe, Wesley J. Everman, Alan C. York, and E. James Dunphy[*]

Recent registrations for dicamba use on dicamba-tolerant soybean will increase the amount of dicamba used and potentially increase incidences of off-target deposition on sensitive crops such as non-dicamba-tolerant soybean. Little information is available on soybean varietal response to dicamba. An experiment was conducted at four sites in 2015 and 2016 to determine response of late-planted maturity group V soybean to sub-lethal rates of dicamba as affected by variety and growth stage at time of dicamba application. Dicamba diglycolamine salt at seven rates ranging from 1.1 to 70 g ae ha^{-1} was applied to four soybean varieties in the V4 or R2 growth stages. Three varieties had a determinate growth habit and one was an indeterminate variety. All rates of dicamba reduced soybean height and yield but a consistent differential variety response was not observed. Averaged over varieties and growth stages at application, soybean injury, height reduction, and yield reduction increased as dicamba rate increased. Injury 14 d after treatment (DAT) ranged from 24 to 76%.

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Height reduction 28 DAT and yield reduction ranged from 12 to 53 and 5 to 81%,
respectively. Yield reduction was correlated with both soybean injury and height reduction.
In contrast to most previous studies, soybean injury, height reduction, and yield reduction
were greater with the V4 application timing. This was likely due to the late planting, with
soybean treated in the V4 stage having less time to recover before initiating reproductive
growth. No consistent response was noted between determinate and indeterminate varieties.

Nomenclature: Dicamba; soybean, *Glycine max* (L.) Merr.

Keywords: Growth stage, height reduction, soil types, variety response.

Dicamba, a benzoic acid herbicide commercialized in the mid-1960's, is commonly used

to control dicot weeds in corn (*Zea mays* L.), sorghum [*Sorghum bicolor* (L.) Moench],

wheat (*Triticum aestivum* L.), pastures, and turf (Behrens et al. 2007; CCME 1999).

Although the mechanism of action of dicamba is not completely understood, the herbicidal

effect has been attributed to an over-induction of the auxin response in susceptible plants

(Grossmann 2000; Kelley and Riechers 2007). Dicot crops such as soybean, cotton

(*Gossypium hirsutum* L.), peanut (*Arachis hypogaea* L.), tobacco (*Nicotiana tabacum* L.),

and most vegetable crops are very sensitive to low rates of dicamba (Al-Khatib and Peterson

1999; Batts 2015; Dittmar et al. 2016; Johnson 2011; Johnson et al. 2012; Kruger et al. 2012;

Mohseni-Moghadam and Doohan 2015; Mohseni-Moghadam et al. 2016; Wall 1994).

Commercial sales of dicamba-tolerant soybean and cotton began in 2015 following

USDA-APHIS deregulation (USDA-APHIS 2015). Certain formulations of dicamba were

registered for use on dicamba-tolerant soybean and cotton beginning with the 2017 season

(Anonymous 2017a,b,d,e). This technology will give growers another tool to aid in

management of herbicide-resistant weeds (Byker et al. 2013; Cahoon et al. 2015; Johnson et

al. 2010; Vann et al. 2017; Vink et al. 2012). At the same time, this technology will enable

use of dicamba on much more land and over a greater portion of the growing season, thus

increasing the risk of injury to dicamba-sensitive crops.

Dicamba injury to susceptible crops due to spray drift, volatilization, and sprayer

contamination has often been observed (Behrens and Lueschen 1979; Boerboom 2004; Egan

et al. 2014; Soltani et al. 2016; Wax et al. 1969). Low rates of dicamba cause crinkling,

cupping, and chlorosis of terminal leaves and reductions in plant height (Andersen et al.

2004; Auch and Arnold 1978; Griffin et al. 2013; Johnson et al. 2012; Robinson et al. 2013; Wax et al. 1969; Weidenhamer et al. 1989). At higher rates, leaf necrosis and swelling and cracking of stems can occur along with death of the terminal bud. Death of the terminal bud and release from apical dominance leads to increased branching from cotyledonary and unifoliate leaf axils resulting in bushy plants. The new formulations of dicamba registered specifically for dicamba-tolerant cotton and soybean (Anonymous 2017a,b,d,e) are purported to have very low volatility (Rhodes et al. 2016; Robertson et al. 2014) although little peer-reviewed data are available to support the claims under field conditions. However, spray drift and contaminated sprayers will continue to be an avenue for exposure of sensitive crops to dicamba (Bode and Zain 1987; Boerboom 2004; Maybank et al. 1978).

A number of studies have been conducted to document the effect of simulated dicamba drift onto soybean. Injury from low rates of dicamba has consistently been observed but results have varied among the studies. In addition to dicamba rate, the extent of the injury and the resulting impact on yield has been affected by soybean growth stage at time of exposure. Griffin et al. (2013) reported greater visible injury 14 DAT with dicamba applied to V3 to V4 stage soybean compared with soybean in the R1 stage but greater height reduction 28 DAT with R1 stage application. Solomon et al. (2014) observed greater injury 14 DAT with dicamba applied to V3 stage soybean compared with soybean in the R2 stage, but the opposite was observed with injury ratings taken 28 DAT. Height reduction 14 DAT was greater with the V3 application whereas no differences in height 28 DAT were observed with the two application timings. Robinson et al (2013) noted greater visible injury 14 DAT with dicamba applied to soybean in the V2 than R2 stage, but injury was greater 28 DAT

with R2 stage application. Plant height, recorded late in the season, was reduced more with the R2 stage application. In most studies, yield reduction has been greater with dicamba application at reproductive stages compared with vegetative stages (Auch and Arnold 1978; Griffin et al 2013; Robinson et al 2013; Solomon et al 2014; Wax et al. 1969). However, Kelley et al. (2005) reported 7% yield reduction from dicamba at 0.56 g ha^{-1} applied to V3 stage soybean but no yield reduction with the same rate of dicamba applied to V7 or R2 stage soybean. Dicamba at 5.6 g ha^{-1} reduced yield 6, 12, and 7% when applied to V3, V7, and R2 stage soybeans, respectively.

In addition to growth stage at time of exposure, other factors also seem to impact soybean injury from dicamba. Greater yield reductions have been reported in drier growing seasons (Andersen et al. 2004; Auch and Arnold 1978; Robinson et al. 2013; Weidenhamer et al. 1989). Al-Khatib and Peterson (1999) observed increased soybean sensitivity to dicamba with high temperatures at time of exposure. Kelley et al. (2005) reported synergistic interactions between dicamba at 5.6 g ha^{-1} and normal use rates of glyphosate, imazethapyr, imazamox, and fomesafen. Thus, the impact of dicamba residue in spray equipment could be enhanced when the equipment is used to apply herbicides to non-dicamba-tolerant soybean. A number of adjuvants are approved for mixing with dicamba applied to dicamba-tolerant crops (Anonymous 2017c,f). Andersen et al. (2004) reported that crop oil concentrate mixed with dicamba greatly increased soybean response. Dicamba at 5.6 g ha^{-1} injured soybean 40 to 45% while dicamba plus the adjuvant caused 90% injury.

Differential variety responses to dicamba have been observed in wheat (*Triticum aestivum*) and barley (*Hordeum vulgare*) (Lemerle et al. 1986; Schroeder and Banks 1989).

Differential soybean variety response has been observed with non-auxin herbicides (Hardcastle 1974; Hulting et al. 2001; Miller et al. 2012; Taylor-Lovell et al. 2001) and with 2,4-D and 2,4,5-T (Fribourg and Johnson 1955). It is therefore not unreasonable to expect there might be a variety response to dicamba but few studies have addressed this. Auch and Arnold (1978) evaluated the response of five soybean varieties to dicamba at 28 g ha^{-1} applied at the early bloom, mid-bloom, or early pod stages. Yields were not reduced with any variety with dicamba applied at the early pod stage. Yields for three and four varieties were reduced when dicamba was applied at the mid-bloom and early bloom stages, respectively. With the early bloom application, yield of the varieties Harcor and Amsoy 71 were reduced 26 to 30% while yield of the varieties Corsoy and SRF-200 were reduced 40 to 49%. Yield of the variety Wells was not affected. However, when dicamba was applied at mid-bloom, yield of Wells was reduced 39% compared with 24 to 29% reduction with SRF-200 and Amsoy 71. Response to dicamba may vary between cultivars with determinate versus indeterminate growth habits. Weidenhamer et al. (1989) compared the response of Elf, a determinate variety, and Williams, an indeterminate variety, to dicamba applied at the pre-bloom and mid-bloom stages. Although statistically valid comparisons could not be made, it appeared that a greater yield reduction occurred with Williams than with Elf, especially with application at the mid-bloom stage. Height reduction was also greater with Williams than Elf with dicamba applied at the mid-bloom stage. A response to dicamba may depend on an interaction between growth habit and growth stage at time of exposure. Wax et al. (1969) suggested that determinate cultivars may be more sensitive to dicamba during

vegetative growth while indeterminate cultivars may be more sensitive during reproductive stages.

In light of very little information available on varietal response to dicamba, we conducted an experiment to determine the response of one indeterminate and three determinate soybean varieties to a range of dicamba rates simulating spray drift or sprayer contamination. A secondary objective was to determine the effect of soybean growth stage at time of dicamba exposure and soil types.

Materials and Methods

The experiment was conducted at the Upper Coastal Plain Research Station (35.89°N, -77.68°W) near Rocky Mount, NC in 2015, the Peanut Belt Research Station (36.13°N, -77.17°W) near Lewiston, NC in 2016, and the Caswell Research Farm (35.27°N, -77.62°W) near Kinston, NC in 2015 and 2016. Soils are described in Table 1. Each location was in a conventional tillage system. Plots were four rows spaced 91-cm apart by 9 m long with the center two rows treated leaving two border rows between treatments. Weed control in soybean was achieved with flumioxazin (Valor SX, Valent U.S.A., Walnut Creek, CA) at 71 g ai ha^{-1} applied PRE followed by glyphosate potassium salt (Roundup PowerMAX, Monsanto Co., St. Louis, Mo) at 1260 g ae ha^{-1} or glufosinate-ammonium (Liberty 280 SL, Bayer CropScience, Research Triangle Park, NC) at 590 g ai ha^{-1} applied twice POST to the appropriate varieties.

Treatments, replicated four times, included a factorial arrangement of four soybean varieties, eight rates of dicamba, and two soybean growth stages at time of dicamba

application in a randomized complete block design. Soybean varieties AG5935 (Monsanto Co., St. Louis, MO), LL 513N (Southern States Cooperative, Richmond, VA), P 5610 RY (Progeny Ag Products, Wynne, AR), and S56RY84 (Crop Production Services, Loveland, CO) were planted at 350,000 seed ha^{-1} with a cone planter on dates in Table 2. These four varieties were selected to represent the range in response to dicamba observed in a preliminary screen of 42 commercial varieties. Dicamba diglycolamine salt (Clarity herbicide, BASF Corporation, Research Triangle Park, NC) was applied to soybean at either the V4 (three completely unrolled trifoliate leaves) or the R2 (full bloom) growth stage. Dicamba application rates were 0, 1.1, 2.2, 4.4, 8.8, 17.5, 35, and 70 g ha^{-1}. These rates represent 0.2 to 12.5% of the recommended rate of 560 g ha^{-1} applied to dicamba-tolerant soybean (Anonymous 2017b,e). Dicamba was applied using a CO_2-pressurized backpack sprayer equipped with flat-fan nozzles (XR11002 extended range flat-spray tips, TeeJet Technologies, Wheaton, IL) calibrated to deliver 140 L ha^{-1} at 170 kPa.

Rainfall was recorded at each location (Table 3). Crop injury was visually estimated 7, 14, and 28 DAT using a scale of 0 (no injury) to 100% (complete death). The injury estimate accounted for the symptomology typical of auxin herbicides (leaf cupping, petiole epinasty, stem swelling, terminal bud death, tissue necrosis). Soybean height was recorded 14 and 28 DAT by randomly selecting four plants from each plot and measuring from the soil surface to the terminal bud. Soybean was mechanically harvested on dates in Table 2 and yields were adjusted to 13% moisture. Soybean height and yield were converted to percent reduction relative to appropriate non-treated checks. Data, excluding non-treated checks, were subjected to ANOVA using the GLM Procedure in SAS 9.3 (SAS Institute Inc., Cary, NC)

and means were separated using Fisher's Protected LSD at p= 0.05. Replications were considered random effects while dicamba rate, growth stage, variety and locations were fixed effects. A separate analysis compared the height and yield of soybean with all dicamba-containing treatments to that of the no-dicamba checks using Dunnett's procedure (Dunnett 1955). Regression analysis was conducted to determine the relationship between dicamba rate and each dependent variable using Pearson's correlation in SAS 9.3.

Results and Discussion

Soybean experienced little to no drought stress at any location (Table 3). Yield at Kinston in 2015 (Table 4) exceeded the North Carolina average yield of 2550 kg ha^{-1} (USDA-NASS 2016). Yields at the other locations were less than the state average yield. This can be attributed largely to the late planting dates (Table 2). Soybean yield in the Mid-Atlantic region of the United States decreases quadratically as planting date is delayed from early June to late July (Parvej et al. 2016). Lower yield with late planting has been attributed to lack of sufficient vegetative growth and failure to intercept 95% of the sunlight by the R1 growth stage (Ball et al. 2000; Egli 1988; Herbert and Litchfield 1984). Soybean in this experiment was well into a reproductive stage when height was recorded 28 d after the V4 application, and height of soybean at that time was less than the target of 91 cm with the canopy closed (Table 4). The lowest yield was obtained at Kinston in 2016. Excessive rainfall from a hurricane during the fourth week after R2 application damaged soybean at this location.

Effect of Dicamba Rate.

A strong dicamba rate response was observed for soybean injury, height reduction, and yield reduction (Tables 5 and 6). There were no dicamba rate by variety interactions for any variable recorded. Additionally, there were no interactions for yield with dicamba rate and any other factor. Interactions of dicamba rate by soil type, dicamba rate by year, and dicamba rate by soil type by year were significant for injury but generally not for height reduction and yield reduction. Dicamba rate by growth stage interactions were significant only for injury at 7 and 14 DAT and height reduction 28 DAT. The mean square for dicamba rate for all variables was at least 31 times greater than for any interactions with rate, hence attention was focused on the dicamba rate main effect.

Similar to results in other recent studies (Griffin et al. 2013; Johnson et al. 2012; Robinson et al. 2013; Soltani et al. (2016), soybean injury, height reduction, and yield reduction increased with increasing dicamba rates. Averaged over varieties, growth stages, years, and soil types, soybean was injured 19 to 60% at 7 DAT as dicamba rate increased from 1.1 to 70 g ha^{-1} (Figure 1). Greater injury was observed at 14 DAT and ranged from 24 to 76%. Similar injury was observed at 28 DAT (data not shown). Other researchers also have reported greater dicamba injury 14 DAT compared with 7 DAT but similar injury at 14 and 28 to 30 DAT (Al-Khatib and Peterson 1999; Griffin et al. 2013). Symptoms such as leaf cupping and petiole epinasty were observed with lower dicamba rates while stem swelling and splitting, foliar necrosis, and terminal meristem death were associated with higher dicamba rates. Symptoms were similar to those reported previously (Al-Khatib and Peterson 1999; Griffin et al. 2013; Johnson et al. 2012; Robinson et al. 2013).

Dunnett's test revealed that soybean height 14 and 28 DAT was significantly reduced by all rates of dicamba. Soybean height reduction 14 DAT and 28 DAT ranged from 13 to 46% and 12 to 53%, respectively, as the dicamba rate increased from 1.1 to 70 g ha^{-1} (Figure 2). Al-Khatib and Peterson (1999) observed height reductions 60 DAT of 7 to 79% when dicamba at 5.6 to 56 g ha^{-1} was applied to soybean in the V3 to V4 stage. Weidenhamer et al. (1989) reported 5 to 62% height reduction 70 DAT as the dicamba rate applied to prebloom soybean increased from 1.3 to 80 g ha^{-1}. Griffin et al. (2013) reported height reductions 28 DAT of 3 to 44% when dicamba rates of 4.4 to 70 g ha^{-1} were applied to V3 to V4 stage soybean. With dicamba at 1.1 to 70 g ha^{-1} applied to R1 stage soybean, the same authors observed 2 to 44% height reduction 28 DAT.

Yield reduction ranged from 5% with 1.1 g ha^{-1} dicamba to 81% with 70 g ha^{-1} (Figure 3). Dunnett's test indicated that dicamba at all rates reduced yield. Griffin et al. (2013) reported yield reductions of 4 to 52% and 3 to 72% with dicamba at 4.4 to 70 g ha^{-1} applied to V3 to V4 stage soybean and 1.1 to 70 g ha^{-1} applied to R1 stage soybean, respectively. Al-Khatib and Peterson (1999) reported 2 to 44% yield reduction with dicamba at 5.6 to 56 g ha^{-1} applied to V3 to V4 stage soybean while Anderson et al. (2004) reported twice that amount of yield reduction (14 to 83%) with the same rates of dicamba at a similar growth stage.

Correlations between dicamba injury and yield reduction and between height reduction and yield reduction were significant. For visible injury 7, 14, and 28 DAT, coefficients were 0.68, 0.69, and 0.70, respectively (data not shown). Coefficients for height reduction 14 and 28 DAT and yield reduction were 0.64 and 0.66, respectively (data not shown). Johnson et al. (2012) reported similar correlation coefficients for dicamba injury 7 and 14 DAT and

yield. Correlation coefficients greater than 0.60 would indicate that both visible injury and height reduction could be considered moderate indicators of potential yield loss (Ott and Longnecker 2001).

Effect of Variety and Growth Stage at Time of Dicamba Application.

One objective of this research was to determine if the four varieties respond differently to dicamba. Main effects of varieties were significant for all variables recorded (Table 5 and 6). However, interactions of variety with growth stage at dicamba application, soil type, and years were present.

Variety Response as Affected by Growth Stage at Time of Dicamba Application. A greater overall response to dicamba was noted with the V4 application compared with the R2 application. Averaged over varieties, dicamba rates, soil types, and years, soybean injury, soybean height reduction, and soybean yield reduction were 7 to 12, 17 to 20, and 8%, respectively, greater with the V4 application (Table 7). The effect of growth stage at time of dicamba application on soybean injury has been variable in other studies. Griffin et al. (2013) reported greater injury 14 DAT with dicamba applied to V3 to V4 stage soybean compared with application at the R1 stage. Kelley et al. (2005) and Solomon et al. (2014) also observed greater injury 14 DAT with application at the V3 stage than at the R2 stage. However, in both studies, injury at 28 to 35 DAT was greater with application at the R2 stage. This indicated soybean treated in the vegetative stage recovered more from the initial injury than soybean treated in the reproductive stage. In both studies (Kelley et al. 2005; Solomon et al. 2014), soybean was planted from mid-May to early June. In our experiment, injury was similar at 14 and 28 DAT, indicating little recovery over time. Lack of recovery

in our study may be related to the late planting dates and three of the four varieties being determinate in growth habit. AG5935, S56RY84, and P 5610 RY have a determinate growth habit while LL 513N is considered as indeterminate (Dunphy 2017). With later planting and determinate varieties, the crop would have less time to recover before going into a reproductive stage. Our results for height reduction and yield reduction are in contrast to those from most previously published studies. Auch and Arnold (1978), Griffin et al. (2013), Robinson et al. (2013), Solomon et al. (2014), Soltani et al. (2016), and Wax et al. (1969) all reported greater soybean height reduction and yield reduction with dicamba applied in reproductive stages than vegetative stages. However, Kelley et al. (2005) observed yield reduction by dicamba at 0.56 g ha^{-1} applied to V3 stage soybean but not R2 stage soybean. At higher rates, yield was reduced similarly with dicamba applied to V3 and R2 stage soybean. Smith (1955) reported greater yield loss by 2,4-D, 2,4,5-T, and 2,4,5-TP when application occurred at the V4 stage compared with application at the early bloom stage. Injury and height reduction were not reported in that study.

Variety by year by growth stage interactions were observed (Table 7). Differences in variety response were inconsistent and in most cases small. With application at the V4 stage in 2015, greatest injury 14 and 28 DAT was noted with LL 513N. The variety with the least injury varied between the two rating dates. Height reduction 14 DAT was least with AG5935 but similar with the other three varieties. No differences among varieties were noted for height reduction 28 DAT. Yield of S56RY84 was reduced less than yield of AG5935, but yields were similar with AG5935, P 5610 RY, and LL 513N. In 2016, injury 14 DAT was similar with all varieties receiving dicamba at the V4 stage. Greater injury 28 DAT was

noted with AG5935 and LL 513N. An opposite response was observed with height reduction. Greater height reductions 14 and 28 DAT were noted with S56RY84 and P 5610 RY. Yield reductions were similar with all varieties.

Variety response to dicamba applied at the R2 stage was also variable. In 2015, injury to AG5935 at 14 DAT was 3 to 4% greater than injury with S56RY84 or P 5610 RY but similar to injury with LL 513N (Table 7). At 28 DAT, 3 to 5% less injury was noted with S56RY84 than other varieties. At both evaluations, injury was numerically greater with AG5935 and LL 513N. The same trend was observed with height reduction, with the greatest height reduction occurring with LL 513N. Height reduction of AG5935 was numerically greater than height reduction of S56RY84 or P 5610 RY. In 2016, the greatest injury was noted with AG5935 while LL 513N sustained the least injury. AG 5935 was injured 10 to 17% more than LL 513N. Height reductions also were greatest with AG5935 but in contrast to the injury ratings where least injury was observed with LL 513N, height reductions were similar with AG 5935 and LL 513N at 14 DAT. At 28 DAT, height reduction was greatest with AG 5935, intermediate with LL 513N, and least with S56RY84 and P 5610 RY. The greatest yield reduction occurred with AG 5935 in 2016 whereas that variety sustained the least yield reduction in 2015.

Growth in height continues after an indeterminate variety of soybean enters the reproductive stage (Bernard 1972). Thus, one might expect dicamba applied during reproductive stages to cause a greater height reduction for an indeterminate variety than for determinate varieties where growth in height mostly ceases once the crop enters the reproductive stage. This was observed with R2 stage applications in 2015 where height

reduction of LL 513N, an indeterminate variety, was greater that the reduction with other varieties (Table 7). However, the response was less clear in 2016. Height reduction due to dicamba applied at R2 in 2016 was greatest with AG5935, intermediate with LL 513N, and least with S56RY84 and P 5610RY.

Variety Response as Affected by Soil Type. Averaged over varieties, growth stages at dicamba application, years, and dicamba rates, soybean injury 28 DAT and soybean yield reduction were somewhat greater on the Rains soil while height reductions were similar on the two soils (Table 8). However, a variety by year by soil type interaction was observed. Within this interaction, the variety response was inconsistent. On the Rains soil, greatest injury and greatest height reduction 28 DAT were noted with AG 5935 and LL 513N in 2015. In 2016, greatest injury on the Rains soil was noted with AG5935 and S56RY84. However, height reductions did not correlate well with injury. No differences among varieties were noted for height reduction 28 DAT. Overall, yield reduction on the Rains soil was greater in 2016 (50%) than in 2015 (40%). Within years, there were no differences among varieties in 2016. In 2015, yield reduction of AG5935 exceeded that of P 5610 RY.

On the Portsmouth soil in 2015, differences in injury among varieties were minor but the least injury was generally observed with S56RY84 (Table 8). Height reductions were similar with all varieties. The yield reduction for P 5610 RY was greater than for S56RY84 while the yield reduction was similar for the other three varieties. On the Portsmouth soil in 2016, injury was greatest with AG5935, intermediate with S56RY84 and P 5610 RY, and least with LL 513N. Height reductions were similar with all varieties. The greatest yield reduction was noted with AG5935 while yields of the other three varieties were similar.

Based on the various interactions observed, growth stage at time of dicamba application and soil type may play a role in determining the extent of injury and yield loss resulting from dicamba exposure. With the four varieties evaluated, there appears to be no clear varietal response to dicamba. A strong response to dicamba rate was observed but no variety by dicamba interaction was observed for any variable (Tables 5 and 6). When averaged over dicamba rates, the effect of variety within three-way interactions of variety by year by growth stage at dicamba application and variety by year by soil type was inconsistent and usually small.

The greater impact of dicamba exposure at the V4 growth stage compared with the R2 stage in this experiment (Table 7), in contrast to most previous experiments, may have been related to the late planting dates. Soybean in this experiment was planted from June 27 to July 15 (Table 2). With late planting, determinate varieties have less time to recover from dicamba exposure at early vegetative stages before initiating reproductive growth. In that situation, the plant would produce fewer nodes and less foliage to support reproductive growth, thus explaining the greater yield impact with the V4 dicamba application. However, we observed no consistent differences between an indeterminate variety and three determinate varieties. Additional research is needed to determine the impact of planting date and growth stage at time of exposure on soybean response to drift rates of dicamba. Regardless of inconsistent responses to varieties, growth stages at time of exposure, and soil types, this experiment, like many others, demonstrated the extreme sensitivity of non-dicamba-tolerant soybean to dicamba and underscores the need for good stewardship practices to avoid off-target deposition of dicamba.

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Table 1. Soil characteristics at experiment sites.

Location	Year	Soil series	Textural classification	Soil subgroup	Soil pH[a]	Humic matter[a]
						%
Kinston	2015	Portsmouth	Loam	Typic Umbraquults	6.2	2.47
Rocky Mount	2015	Rains	Sandy loam	Typic Paleaquults	5.8	0.60
Kinston	2016	Portsmouth	Loam	Typic Umbraquults	6.2	2.37
Lewiston	2016	Rains	Sandy loam	Typic Paleaquults	5.9	0.92

[a] Soil pH and humic matter determined by the North Carolina Department of Agriculture and Consumer Services,

Agronomic Division. Humic matter was determined photometrically according to Mehlich (1984).

Table 2. Soybean planting and harvest dates and dicamba application dates.

Location	Year	Planting date	Dicamba application date		Harvest date
			V4	R2	
Kinston	2015	July 15	August 10	August 30	December 12
Rocky Mount	2015	July 9	August 4	August 25	December 9
Kinston	2016	June 27	July 23	August 12	December 1
Lewiston	2016	June 30	July 29	August 15	November 16

Table 3. On-site rainfall at experiment locations.

Time[a]	Kinston 2015	Rocky Mount 2015	Kinston 2016	Lewiston 2016
	cm			
2 wk before V4	0	3.2	5.5	8.7
1 wk before V4	0	0.2	3.3	0.1
1 wk after V4	4.5	2.9	0	5.0
2 wk after V4	1.2	0.7	3.5	2.0
1 wk before R2	0.4	1.8	3.7	3.3
1 wk after R2	0.7	2.9	0.2	2.0
2 wk after R2	2.0	4.9	4.6	0
3 wk after R2	0	3.3	3.3	14.6
4 wk after R2	3.7	0	13.0	1.2
5 wk after R2	11.9	5.3	0	0
6 wk after R2	2.7	15.1	9.8	24.7

[a] Time in relation to dicamba applications at V4 and R2 soybean growth stages.

Table 4. Height and yield of non-treated soybean.

Location	Year	Soybean height				Soybean yield
		V4 application		R2 application		
		14 DAT[a]	28 DAT	14 DAT	28 DAT	
		cm				kg ha^{-1}
Kinston	2015	48	75	74	75	2560
Rocky Mount	2015	31	50	53	53	1690
Kinston	2016	31	60	68	73	1350
Lewiston	2016	29	53	67	69	1680

[a] Abbreviation: DAT, days after treatment.

Table 5. Analysis of variance for soybean injury.[a]

Source	df	7 DAT		14 DAT		28 DAT	
		Mean square	p-value	Mean square	p-value	Mean square	p-value
Soil	1	60	0.1177	772	<0.0001	1927	<0.0001
Year	1	1542	<0.0001	87	0.0844	28	0.3336
Soil*Year	1	297	0.0005	56	0.1634	9	0.5789
Variety (Var)	3	185	<0.0001	469	<0.0001	1017	<0.0001
Soil*Var	3	110	0.0037	34	0.3172	31	0.3677
Year*Var	3	299	0.0001	412	<0.0001	755	<0.0001
Soil*Var*Year	3	76	0.0260	39	0.2535	85	0.0348
Rate	6	27804	<0.0001	43113	<0.0001	47888	<0.0001
Soil*Rate	6	149	<0.0001	403	<0.0001	994	<0.0001
Year*Rate	6	203	<0.0001	103	0.0019	170	<0.0001
Soil*Year*Rate	6	316	<0.0001	354	<0.0001	272	<0.0001
Var*Rate	18	27	0.3536	29	0.4772	27	0.5393
Soil*Var*Rate	18	20	0.6721	19	0.8542	30	0.4360
Year*Var*Rate	18	46	0.0147	42	0.1057	39	0.1613
Soil*Year*Var*Rate	18	30	0.2318	20	0.8133	25	0.6286

Table 5 (Continued)

Growth stage (GS)	1	4139	<0.0001	31355	<0.0001	9613	<0.0001
Soil*GS	1	4282	<0.0001	4462	<0.0001	6012	<0.0001
Year*GS	1	4153	<0.0001	9	0.5787	64	0.1399
Soil*Year*GS	1	158	0.0110	105	0.0577	90	0.0803
Var*GS	3	205	<0.0001	552	<0.0001	646	<0.0001
Soil*Var*GS	3	112	0.0033	367	<0.0001	366	<0.0001
Year*Var*GS	3	20	0.4819	77	0.0466	127	0.0050
Soil*Year*Variety*GS	3	27	0.3494	61	0.0971	141	0.0027
Rate*GS	6	257	<0.0001	230	<0.0001	28	0.4573
Soil*Rate*GS	6	257	<0.0001	568	<0.0001	850	<0.0001
Year*Rate*GS	6	76	0.0050	400	<0.0001	227	<0.0001
Soil*Year*Rate*GS	6	27	0.3512	13	0.8308	11	0.8893
Var*Rate*GS	18	35	0.1071	40	0.1344	35	0.2582
Soil*Var*Rate*GS	18	28	0.2905	51	0.0258	45	0.0792
Year*Var*Rate*GS	18	19	0.7098	40	0.1304	42	0.1053
Soil*Year*Var*Rate*GS	18	16	0.8373	46	0.0560	48	0.0477

[a] Abbreviations: df, degrees of freedom; DAT, days after treatment.

Table 6. Analysis of variance for soybean height reduction and yield reduction.[a]

| | | Height reduction | | | | Yield reduction | |
| | | 14 DAT | | 28 DAT | | | |
Source	df	Mean square	p-value	Mean square	p-value	Mean square	p-value
Soil	1	19	0.6852	0.65	0.9426	2665	0.0497
Year	1	1090	0.0020	49	0.5357	472	0.4085
Soil*Year	1	18	0.6890	1195	0.0022	18054	<0.0001
Variety (Var)	3	845	<0.0001	613	0.0024	2374	0.0165
Soil*Var	3	1083	<0.0001	191	0.2116	3074	0.0041
Year*Var	3	849	<0.0001	578	0.0036	679	0.3996
Soil*Var*Year	3	1646	<0.0001	648	0.0017	4636	0.0002
Rate	6	18332	<0.0001	26255	<0.0001	106317	<0.0001
Soil*Rate	6	41	0.9020	99	0.5862	283	0.8720
Year*Rate	6	459	0.0005	146	0.3306	1323	0.0753
Soil*Year*Rate	6	94	0.5490	187	0.1840	724	0.3914
Var*Rate	18	81	0.8026	73	0.9154	911	0.1672
Soil*Var*Rate	18	97	0.6352	136	0.3779	935	0.1468
Year*Var*Rate	18	110	0.4977	101	0.7047	603	0.6108

Table 6 (Continued)

Soil*Year*Var*Rate	18	40	0.9945	34	0.9991	859	0.2180
Growth stage (GS)	1	68081	<0.0001	89157	<0.0001	8641	0.0004
Soil*GS	1	823	0.0072	1867	0.0001	27196	<0.0001
Year*GS	1	3589	<0.0001	746	0.0154	1376	0.1582
Soil*Year*GS	1	224	0.1599	3451	<0.0001	1047	0.2182
Var*GS	3	2666	<0.0001	3725	<0.0001	1238	0.1468
Soil*Var*GS	3	481	0.0055	382	0.0294	263	0.7663
Year*Var*GS	3	913	<0.0001	2181	<0.0001	3930	0.0007
Soil*Year*Variety*GS	3	74	0.5836	611	0.0025	1482	0.0928
Rate*GS	6	201	0.1031	499	0.0007	894	0.2561
Soil*Rate*GS	6	144	0.2678	264	0.0528	789	0.3348
Year*Rate*GS	6	591	<0.0001	829	<0.0001	201	0.9407
Soil*Year*Rate*GS	6	140	0.2850	82	0.6925	1281	0.0857
Var*Rate*GS	18	57	0.9555	32	0.9994	787	0.3061
Soil*Var*Rate*GS	18	36	0.9971	99	0.7204	813	0.2722
Year*Var*Rate*GS	18	28	0.9995	42	0.9963	223	0.9967
Soil*Year*Var*Rate*GS	18	58	0.9540	63	0.9605	561	0.6848

[a] Abbreviations: df, degrees of freedom; DAT, days after treatment.

Table 7. Soybean variety response to dicamba as affected by year and growth stage at time of dicamba application.[a]

Variety	Year	Growth stage	Injury		Height reduction		Yield reduction
			14 DAT[b]	28 DAT	14 DAT	28 DAT	
			%				
AG5935	2015	V4	50 bc	45 bc	38 bc	44 ab	54 a
S56RY84	2015	V4	48 c	42 d	43 a	43 abc	43 b-e
P 5610 RY	2015	V4	43 d	45 bc	41 ab	40 bc	46 a-d
LL 513N	2015	V4	53 a	49 a	44 a	40 bc	46 a-d
AG5935	2016	V4	51 ab	48 a	28 d	33 d	50 ab
S56RY84	2016	V4	51 ab	45 bc	40 ab	46 a	42 b-f
P 5610 RY	2016	V4	49 bc	44 c	38 bc	45 a	45 a-d
LL 513N	2016	V4	51 ab	47 ab	35 c	39 c	43 b-e
AG5935	2015	R2	40 e	41 de	19 fg	19 f	34 f
S56RY84	2015	R2	37 f	36 hi	15 g	16 f	36 ef
P 5610 RY	2015	R2	36 fg	39 efg	18 fg	15 f	44 b-e
LL 513N	2015	R2	38 ef	40 def	29 d	30 d	40 c-f

Table 7 (Continued)

AG5935	2016	R2	44 d	47 ab	25 de	30 d	47 abc
S56RY84	2016	R2	38 ef	37 gh	19 fg	16 f	28 g
P 5610 RY	2016	R2	38 ef	38 fgh	18 fg	18 f	40 c-f
LL 513N	2016	R2	34 g	34 i	22 ef	24 e	37 def
Main effect of growth stage[c]		V4	50*	46*	38*	41*	46*
		R2	38	39	21	21	39

[a] Data averaged over dicamba rates and soil types. Means within a column followed by the same letter are not different at P = 0.05 according to Fisher's Protected LSD.

[b] Abbreviation: DAT, days after treatment.

[c] An asterisk denotes a significant difference between growth stages at P = 0.05.

Table 8. Soybean variety response to dicamba as affected by year and soil type.[a]

			Injury		Height reduction		Yield
Variety	Year	Soil type	7 DAT[b]	28 DAT	14 DAT	28 DAT	reduction
					%		
AG5935	2015	Rains	36 c	45 bc	28 bcd	32 bc	43 bc
S56RY84	2015	Rains	33 ef	40 fgh	27 cd	26 d	40 cd
P 5610 RY	2015	Rains	32 f	43 cde	27 cd	24 d	38 def
LL 513N	2015	Rains	35 cd	47 ab	44 a	37 a	38 cde
AG5935	2015	Portsmouth	34 de	41 efg	30 bcd	32 bc	44 abc
S56RY84	2015	Portsmouth	33 ef	38 h	31 bc	32 bc	39 cd
P 5610 RY	2015	Portsmouth	35 cd	40 fgh	32 b	31 bc	51 ab
LL 513N	2015	Portsmouth	36 c	42 def	30 bcd	33 ab	47 abc
AG5935	2016	Rains	39 ab	48 a	26 d	32 bc	45 abc
S56RY84	2016	Rains	40 a	44 cd	32 b	34 ab	51 ab
P 5610 RY	2016	Rains	36 c	41 efg	29 bcd	33 ab	52 ab
LL 513N	2016	Rains	36 c	42 def	28 bcd	32 bc	50 ab

Table 8 (Continued)

AG5935	2016	Portsmouth	38 b	47 ab	31 bc	31 bc	53 a
S56RY84	2016	Portsmouth	36 c	39 gh	28 bcd	28 cd	25 f
P 5610 RY	2016	Portsmouth	36 c	40 fgh	27 cd	31 bc	33 def
LL 513N	2016	Portsmouth	34 de	30 i	29 bcd	31 bc	29 ef
Main effect of soil type[c]		Rains	36	44*	30	31	45*
		Portsmouth	35	40	30	31	40

[a] Data averaged over dicamba rates and growth stages. Means within a column followed by the same letter are not different at $P = 0.05$ according to Fisher's Protected LSD.

[b] Abbreviation: DAT, days after treatment.

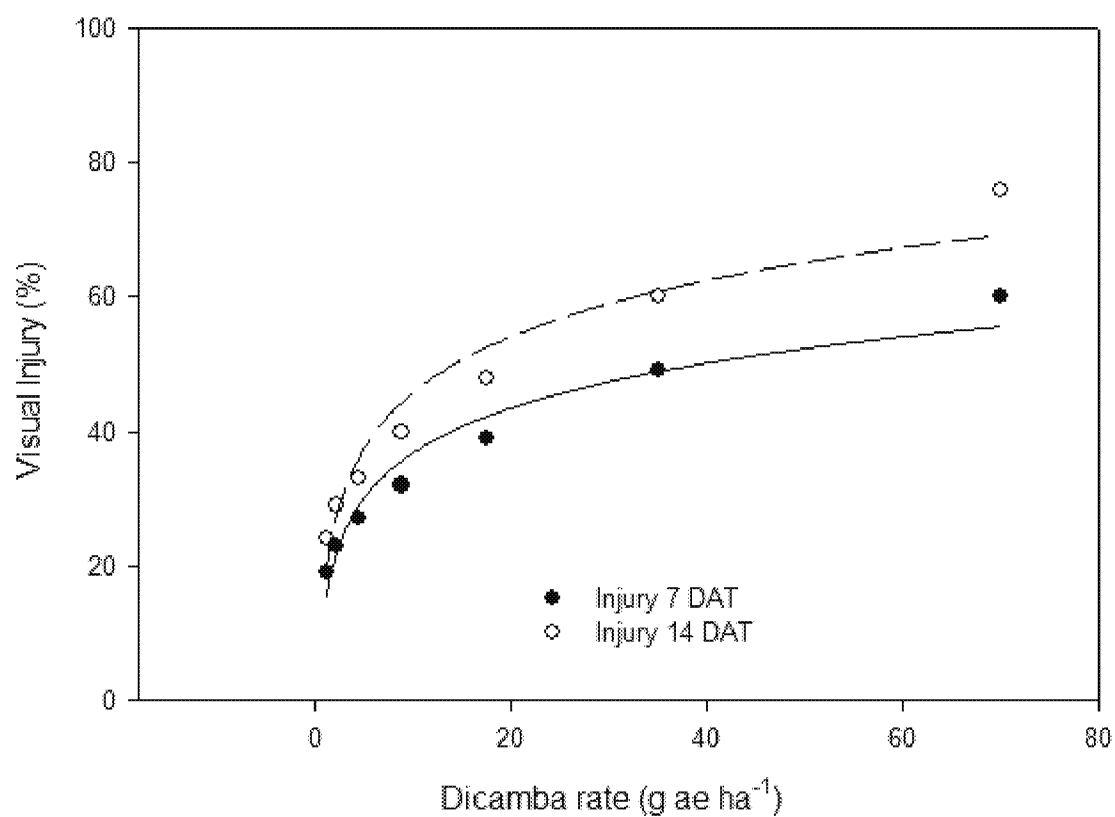[c] An asterisk denotes a significant difference between soil types at $P = 0.05$.

Figure 1. Soybean injury as affected by dicamba rate (1.1-70 g ae ha^{-1}) 7 and 14 days after treatment (DAT). Predicted response 7 DAT can be described as Y= 20.57+1.16x-0.009x^2, r^2=0.98. Predicted response 14 DAT can be described as Y=26+1.36x-0.009x^2, r^2=0.98.
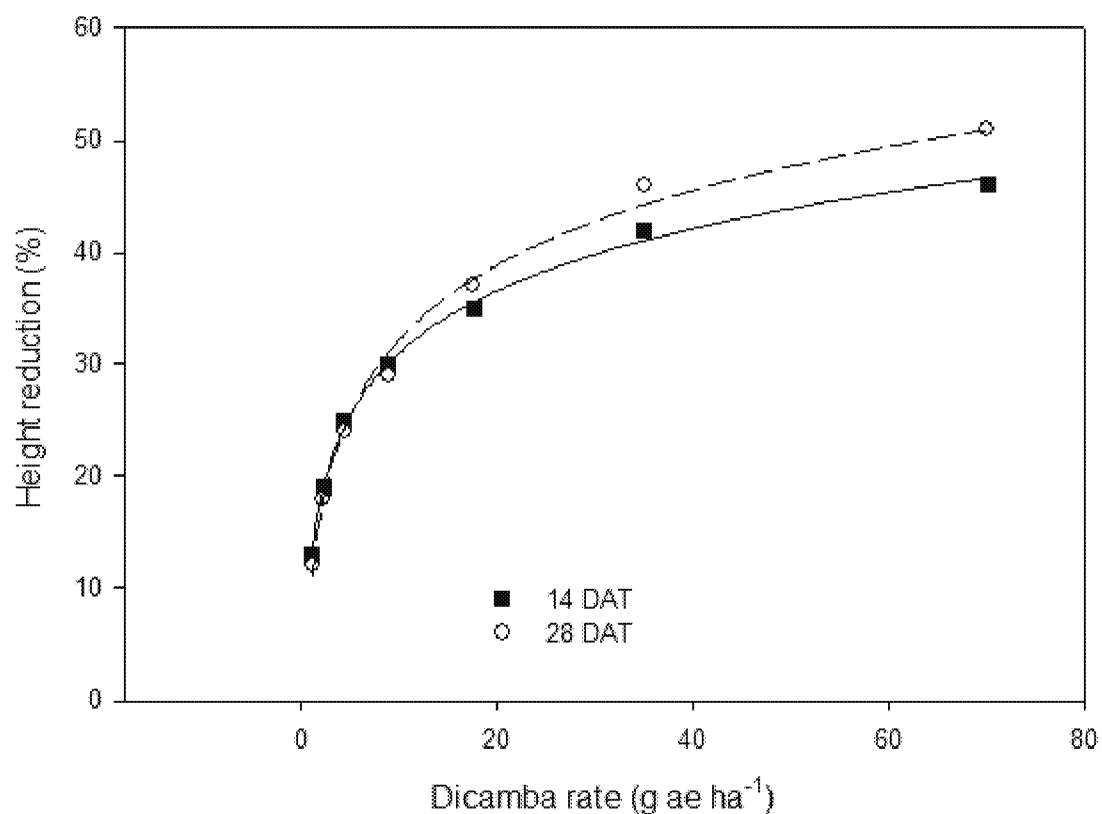
Figure 2. Soybean height reduction as affected by dicamba rate (1.1-70 g ae ha^{-1}) 14

and 28 days after treatment (DAT). Predicted response 14 DAT is described by

Y=12.6+8*ln(abs(x)), r^2= 0.99. Predicted response 28 DAT is described by

Y=9.9+9.8*ln(abs(x)), r^2=0.99.
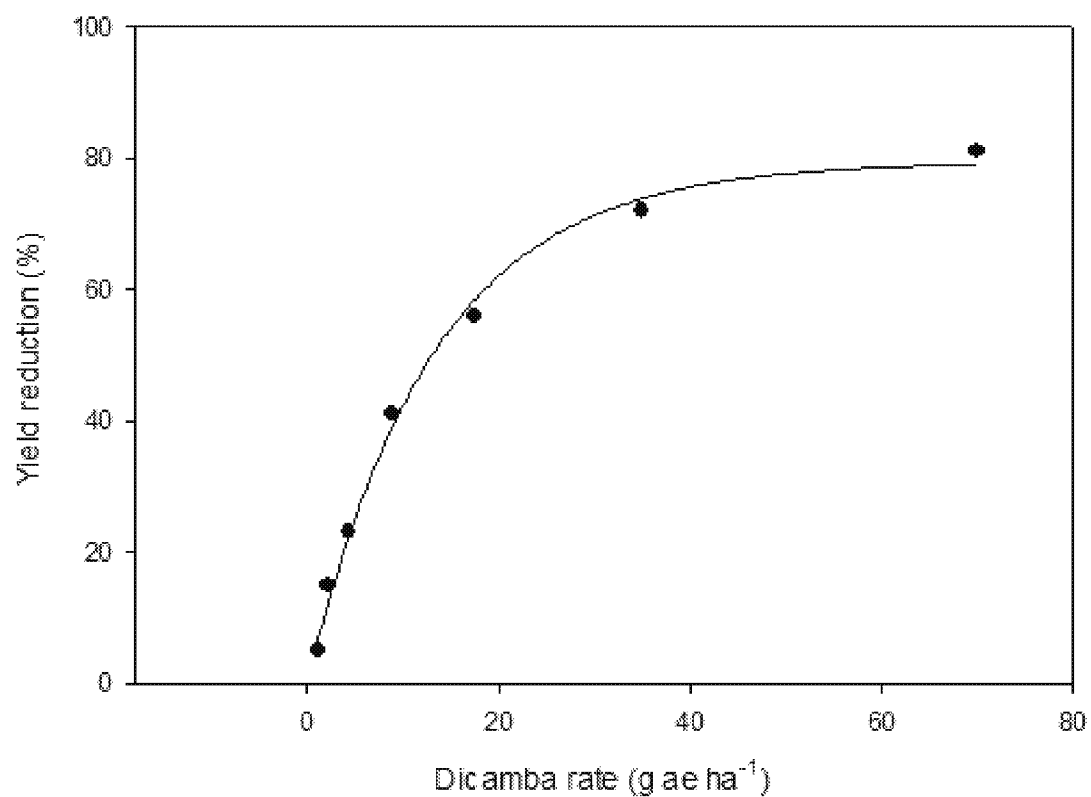
Figure 3. Soybean yield reductions as affected by dicamba rate (1.1-70 g ae ha^{-1}).

Predicted yield reduction can be described as Y=-0.19+19*ln(abs(x)), r^2=0.99.

Chapter III

Effects of Simulated Dicamba Drift on Maturity Group VI Soybean

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Abstract

Dicamba-tolerant crop varieties have the potential to become utilized in North

Carolina as a tool to control glyphosate-resistant weeds. Due to North Carolina's diverse

agricultural landscape, there is concern about potential off-target movement of this broadleaf

herbicide to sensitive crops. Previous research has determined that soybean cultivars,

commonly glyphosate- or glufosinate-tolerant varieties, are highly sensitive to

dicamba. Tank contamination, wind drift, and volatility of dicamba have been shown to

cause injury and reduce soybean yields. To date, there has been little information reported on

soybean varietal responses to sub-lethal doses of dicamba, therefore, the objective of this

study was to evaluate the effects of sub-lethal rates of dicamba on five maturity group VI

soybean cultivars at the vegetative and reproductive growth stages. Effects of dicamba were

determined by collecting visual injury ratings, height reductions, and yield. The experiment

was conducted in Lewiston and Kinston, NC in 2015 and Rocky Mount, NC in 2016, with a

total of three site years. These locations were chosen to represent different growing

environments, with Kinston having a Portsmouth loam soil and Lewiston and Rocky Mount

having a Rains sandy loam. Five soybean varieties were treated with dicamba at 1.1, 2.2,

4.4, 8.8, 17.5, 35, and 70 g ae ha (1/512 to 1/8 of the labeled use rate for weed control for

dicamba-tolerant soybean) during the V4 or R2 growth stage. Experiments were conducted

using a factorial arrangement of treatments in a randomized complete block design, with

factors being dicamba rate, application timing, and soybean cultivar. Analysis showed a

wide range of visual injury and height reductions 14 and 28 DAT (days after treatment) for

all five varieties. Soybean heights were reduced and injury was greater as dicamba rates

increased. Height reductions 14and 28 DAT (weeks after treatment) were greater for the vegetative growth stage compared to the reproductive stage. Yield reductions ranged from 7 to 67% as dicamba rate increased from 1.1 to70 g ha^{-1}. Analysis revealed a year, soil, variety, and timing interaction for yield reduction. At Rocky Mount and Lewiston, greater yield reduction was observed for the reproductive application but yield reduction due to application timing was not significant for Kinston. Although a variety response was observed for each trial, the results were inconsistent. Generally, variety SS 6810NR2 was more sensitive to dicamba across all years. Injury ratings and height reductions were moderately correlated with yield loss. The correlation coefficient between injury 7, 14 and 28 DAT and yield reduction was greater than 0.6. The correlation coefficient for height reductions 14 and 28 DAT was lower at 0.49 and 0.51, respectively. Not only do these data suggest there may be a varietal response, there are also implications that environmental factors (such as soil type, precipitation, temperature) heavily influence soybean response to dicamba drift. With such variability between environments, yield loss may be difficult to accurately predict. It is important that growers use best management practices if using dicamba or other auxin technologies as a weed control option.

Nomenclature: Dicamba; soybean, *Glycine max* (L.) Merr

Keywords: Off-target movement; contamination; varietal response

Introduction

Synthetic auxin herbicides are the oldest herbicide class dating back to the development of 2,4-D in the 1940's and dicamba in the late 1950's. Dicamba is a synthetic auxin herbicide in the benzoic acid chemical family. Its mode of action is classified as a TIR1 (transport inhibitor response) inhibitor and is group 4 on WSSA herbicide list. Today, there is renewed interest in utilizing auxin herbicides in cropping systems. Current corn (*Zea mays*) and grain sorghum (*Sorghum bicolor*) weed management programs include the older conventional chemistries like dicamba and 2, 4-D to manage glyphosate-resistant weeds such as Palmer amaranth [*Amaranthus palmeri* S. Wats.] and common ragweed [*Ambrosia artemiisifolia* L.]. Although the release of dicamba- and 2,4-D- tolerant crops will allow growers to utilize another mode of action to control these troublesome weeds in cotton and soybean, the use of auxin herbicides in North Carolina's diverse agricultural landscape has raised concerns of off-target movement of these products to sensitive crops, such as non-dicamba-tolerant and non-2,4-D tolerant cotton (*Gossypium hirsutum* L.), peanut (*Arachis hypogaea* L.), tobacco (*Nicotiana tabacum* L.) and soybean.

Because of dicamba's volatile nature (Bunch and Gervais, 2012), much research has focused on quantifying volatility and drift concentrations over the last 50 years. Vapor drift has been quantified at a concentration of 0.56 g ha^{-1}, which is about 1/1000th of a labeled rate of dicamba (Egan and Mortensen 2012). Maybank et al. (1978) calculated downwind drift deposits from unshielded ground sprayers can be 1 to 8%, which is 5.6 to 44.8 g ha^{-1} of dicamba. Quantifying these off-target rates has allowed for more relevant research in understanding how sensitive crops respond to realistic sub-lethal doses of dicamba.

Previous trials have been conducted to observe the effects of dicamba rates on soybean yield and to devise numerical injury and height reduction scales to describe the severity of injury and predict yield loss. Griffin et al. (2013) conducted a field trial by applying 4.4, 8.8, 17.5, 35, 70, 140, and 280 g ha^{-1} (1/128 to 1/2 of the recommended use rate of 560 g ha^{-1}) of dicamba at the 2- to 3- trifoliate and reproductive stage. Visual injury ratings, plant heights, and yield were recorded. When dicamba was applied at 4.4 to 17.5 g ha^{1}, predicted mature height was reduced 3 to 9% and yield was reduced 4 to 15%. Griffin et al. (2013) also reported a greater yield impact for the reproductive growth stage compared to the vegetative stage and concluded that soybean was 2.5 times more sensitive to dicamba in the reproductive stage compared to the vegetative stage. Similarly, researchers in South Dakota reported that soybean was most sensitive to dicamba exposure during the early bloom stage (Auch and Arnold 1978). Soybean heights and yield were significantly reduced when 11 g ha^{-1} was applied during the early bloom stage. While heights were reduced for the vegetative stage, no yield reduction was observed. The authors concluded that one could not always expect yield reduction when plant heights are reduced from dicamba exposure. Johnson et al. (2012) conducted studies to correlate visual injury symptoms with yield as an aid in making additional management decisions concerning dicamba injury on soybeans. Correlations between visual injury 7 and 14 DAT and yield were significant with a correlation coefficient of -0.67 and -0.60, respectively. It was determined that visual injury ratings were a moderate indicator of soybean yield loss.

Dicamba impacts on soybean have been well documented. However, there is little published information examining cultivars' response to sub-lethal rates of dicamba. With

variability in injury, height, and yield reductions reported in previous literature, research should be conducted to address if variety influences the response to dicamba drift. The last soybean variety test examining dicamba effects was published in 1978. Auch and Arnold (1978) evaluated the response of five soybean varieties to dicamba at 28 g ha^{-1} applied at the early bloom, mid-bloom, or early pod stages. Yields were not reduced with any variety with dicamba applied at the early pod stage. Yields for three out of four varieties were reduced when dicamba was applied at the mid-bloom and early bloom stages, respectively. With the early bloom application, yield of the varieties Harcor and Amsoy 71 were reduced 26 to 30% while yield of the varieties Corsoy and SRF-200 were reduced 40 to 49%. Yield of the variety Wells was not affected. However, when dicamba was applied at mid-bloom, yield of Wells was reduced 39% compared with 24 to 29% reduction with SRF-200 and Amsoy 71. These data suggest that when soybean is subjected to sub-lethal doses of dicamba, variety may influence the predicted yield loss. With little information available on varietal response to dicamba, an experiment was conducted to determine the response of five maturity group VI soybean varieties to a range of dicamba rates simulating spray drift or sprayer contamination. A secondary objective was to determine the effect of soybean growth stage at time of dicamba exposure and soil types.

Materials and Methods

An experiment was conducted to evaluate the effects of sub-lethal rates of dicamba on five maturity group VI soybean cultivars at vegetative and reproductive growth stages. The design was a factorial arrangement of 80 treatments in a randomized complete block with four replications and three factors consisting of dicamba rate, soybean cultivar, and soybean growth stage. Trials were conducted at the Caswell Research Farm near Kinston, NC in 2015, the Peanut Belt Research Station near Lewiston-Woodville, NC in 2015, and the Upper Coastal Plains Research Station near Rocky Mount, NC in 2016 which represented three environments. Soil type, planting date, and harvest date for each trial can be found in Table 1. In each trial, five soybean varieties, CZ 6316LL (Bayer Crop Science., Research Triangle Park, NC 27709) DG S64LS95 (Crop Production Services, Loveland, CO 80538), DG S69RY34 (Crop Production Services, Loveland, CO 80538), SH 6515LL (Meherrin Agriculture, Claxton, GA 30417), and SS 6810NR2 (Southern States Cooperative, Richmond, VA 23230), were selected from an initial test (data not shown) and planted using a two-row cone planter at a seeding rate of 350,000 seed ha^{-1} on 91-cm row spacing. After planting, flumioxazin (Valor SX, Valent U.S.A., Walnut Creek, CA 94598) at 71 g ai ha^{-1} was applied PRE followed by two POST applications of glyphosate (Roundup PowerMAX, Monsanto Co., St. Louis, MO 63167) at 1260 g ae ha^{-1} or glufosinate-ammonium (Liberty, Bayer Crop Science., Research Triangle Park, NC 27790) at 590 g ai ha^{-1} to eliminate weed competition.

The diglycolamine (DGA) salt formulation of dicamba (Clarity herbicide, BASF Corporation, Research Triangle Park, NC 27709) was applied to soybean at 1.1, 2.2, 4.4, 8.8,

17.5, 35, and 70 g ha^{-1} (1/512 to 1/8 of the labeled 560 g ha^{-1} use rate for weed control in dicamba-tolerant soybean) when soybeans reached V4 (three completely unrolled trifoliates) or R2 (full bloom) growth stages. A non-treated control was included for each variety. Applications were made using a CO_2-pressurized backpack sprayer equipped with flat-fan nozzles (TeeJet XR 11002 VS, TeeJet Technologies, Springfield, IL 62703) calibrated to deliver 140 L ha^{-1} at 170 kPa. Weather conditions during each application were monitored and recorded using a Kestrel 3000 Pocket Weather Meter (Weather Republic, LLC, 3947 West Lincoln Highway, Suite 304, Downingtown, PA 19335) (Table 2). Plot dimensions were 3.65 m wide by 9 m long and consisted of four rows, with the two center rows being treated and two border rows to minimize cross contamination from herbicide applications. After each application, effects of dicamba were determined by collecting visual injury ratings at 7, 14, and 28 DAT using a scale of 0 (no injury) to 100% (complete death). Symptomology typical of auxin herbicides (leaf cupping, petiole epinasty, stem swelling, terminal bud death, and tissue necrosis) were factored into the injury rating. Soybean height was recorded 0, 14, and 28 DAT by randomly selecting four plants from each plot and measuring from the soil surface to the terminal bud. The treated rows for each plot were mechanically harvested and yields were adjusted to 13% moisture. For ease of comparison, plant heights and yield are expressed as a percent reduction of the respective non-treated check. All data were subjected to ANOVA using the GLM Procedure in SAS 9.3 (SAS Institute Inc., 100 SAS Campus Dr., Cary, NC 27513) and means were separated using Fisher's Protected LSD at p= 0.05. Regression analysis was calculated to determine the relationship between dicamba rate and each dependent variable. The relationship between

visual injury, plant height reduction, and yield loss were examined using Pearson's correlation procedure in SAS. With the objective to understand how much dicamba is required to significantly reduce yield, treatments were compared to the non-treated using Dunnett's test at $\alpha = 0.05$ (Dunnett 1955).

Results and Discussion

Dicamba Rate.

A significant dose response was observed for soybean injury, height reduction, and yield reduction pooled across varieties, application timings, and environments. The mean square for dicamba rate for all variables was at least 20 times greater than any interactions with rate so rate as a main effect will be discussed. Based on Dunnett's test, plant heights and yield were significantly different from the non-treated when dicamba was applied at 1.1 and 2.2 g ha^{-1}, respectively (data not shown). Similar to results in other recent studies (Griffin et al. 2013; Johnson et al. 2012; Robinson et al. 2013; Soltani et al. 2016), soybean injury, height reduction, and yield reduction increased with increasing dicamba rates. As dicamba rate increased from 1.1 to 70 g ha^{-1}, injury 7 DAT ranged from 14 to 53% (Figure 1). An increase in visual injury was noted 14 DAT, ranging from 18 to 66% and by 28 DAT there was little change from 14 DAT (Figure 1). Other researchers also have reported greater dicamba injury 14 DAT compared with 7 DAT but similar injury at 14 and 28 to 30 DAT (Al-Khatib and Peterson 1999; Griffin et al. 2013). Symptoms such as leaf cupping and petiole epinasty were observed with lower dicamba rates while stem swelling and splitting, foliar necrosis, and terminal meristem death were associated with higher dicamba rates.

Dicamba rate, when averaged over soybean variety, application timing, and environment, also significantly affected soybean height and yield. As dicamba rate increased, soybean height reductions 14 DAT increased from 3 to 39%, and by 28 DAT, soybean height reductions ranged from 6 to 42% (Figure 2). Impacts of dicamba were observable through harvest, with yield reductions ranging from 6 to 70% as dicamba rate increased from 1.1 to 70 g ha^{-1} (Figure 3). Researchers in Louisiana reported a 10% yield reduction when dicamba was applied at 4.4 g ha^{-1} to R1 soybean (Griffin et al. 2013). For the current study, when pooled across all varieties, timings, and environments, a yield reduction of 20% was noted for the same rate.

Correlations between injury ratings and yield reduction were significant for dicamba application. For visual injury ratings 7, 14, and 28 DAT, coefficients were 0.57, 0.59, and 0.61, respectively (Table 3). The relationship of height reductions 14 and 28 DAT and yield reduction was not as strong, with correlation coefficients of 0.41 and 0.43, respectively. The relationship of the data implies that as injury and height reductions increase, yield reduction also increases. With a coefficient greater than 0.60, injury ratings 28 DAT are considered a moderate indicator of yield loss (Ott and Longnecker 2001). While the relationship of height reduction and yield reduction is significant, injury ratings may be a stronger indicator than height reductions. Similarly, Johnson et al. (2012) reported a moderate relationship between injury ratings 1 and 2 WAT and soybean yield reduction with correlation coefficients of 0.67 and 0.60, respectively. Weidenhamer et al. (1989) concluded that height reductions are good predictors of yield reductions from dicamba injury.

Environment, Rate, and Application Timing.

A significant interaction of environment, dicamba rate, and application timing was observed for injury 7, 14 and 28 DAT and height reduction 14 and 28 DAT. Soybean injury was evident at all dicamba rates and both application timings for all environments evaluated (Figure 4, 5, and 6). When dicamba was applied at 17.5 g ha^{-1} or less, visual injury ratings 7 DAT were less than 40% regardless of environment (Figure 4). However, differences in injury due to application timing were most evident for all environments at the highest dicamba rate, with greater injury in all environments observed when applications were made to R2 soybeans, regardless of dicamba rate. The injury observed when dicamba was applied to R2 soybeans continued to increase as time after application increased, with greatest injury occurring 28 DAT (Figures 5 and 6). However, when soybeans were treated at V4, injury was similar from 7 to 28 DAT, indicating more recovery compared to those treated at R2. Differences in injury due to application timing 28 DAT were more evident at Rocky Mount when dicamba was applied at 2.2 g ha^{-1} or greater (Figure 6). The greatest injury was observed at Rocky Mount and ranged from 19 to 83% when the application was made to R2 soybean compared to the V4 application which ranged from 17 to 54% (Figure 6). Similarly, injury 28 DAT for Lewiston was generally greater when dicamba was applied to R2 soybeans, which ranged from 18 to 75%, compared to the V4 growth stage which ranged from 5 to 48%. At the Kinston location, when dicamba was applied at 35 g ha^{-1} or less, injury 28 DAT was greater for the V4 application, ranging from 20 to 50%, compared to the R2 application which ranged from 13 to 46%.

Regardless of environment or application timing, height reductions 14 DAT were less than or equal to 20% when dicamba was applied at 2.2 g ha^{-1} or lower (Figure 7). Height reductions 14 DAT were generally greater for V4 applications compared to R2. At Rocky Mount and Kinston, differences in height reduction due to application timing were more evident once dicamba rates increased to 8.8 g ha^{-1} and greater. For Rocky Mount, height reductions were greater for the V4 timing ranging from 37 to 43% compared to the R2 timing which ranged from 22 to 37% (Figure 7). Similarly, at Kinston, greater height reduction was observed for the V4 timing and ranged from 35 to 60% compared to 7 to 17% R2 application. For the Lewiston location, separation between timings was observed at 35 g ha^{-1} and greater, with greater height reduction recorded for the V4 application. Trends in soybean height reductions 28 DAT were similar to 14 DAT (Figure 8). The greatest height reduction 28 DAT was observed after the V4 application in Rocky Mount and Kinston which ranged from 11 to 56% and 11 to 60%, respectively. Height reductions were generally greater for the V4 applications compared to R2 and can be attributed to the determinate nature of the varieties tested. When the reproductive application was made to these determinate varieties, the majority of vegetative growth had been completed which minimized the effect dicamba had on plant heights (Bernard, 1972). Height reduction was generally more severe at Rocky Mount. This may be attributed to an earlier planting date at this location, which allowed non-treated soybeans to reach full height potential by the end of the season, amplifying the effects of sub-lethal dicamba rates on treated plants.

The interaction of environment, dicamba rate, and application timing highlights the variable nature of dicamba and its effects on soybean. Environmental factors such as soil

moisture, ambient temperature, and subsequently growing degree days will have an impact on soybean response to dicamba, regardless of rate.

Rate by Application Timing.

ANOVA revealed a rate by timing interaction, pooled over environment and varieties, for yield reduction. As dicamba rate increased from 1.1 to 70 g ha^{-1}, yield reduction ranged from 0 to 58% for the V4 application and 10 to 80% for the R2 application (Figure 9). These data suggest, when pooled across environments and varieties, that the reproductive stage is more sensitive to dicamba exposure than vegetative growth stages. These data agree with the current consensus that yield reduction is greater with dicamba application at reproductive stages compared with vegetative stages (Auch and Arnold 1978; Robinson et al 2013; Solomon et al 2014; Wax et al. 1969). Griffin et al concluded that soybean in the reproductive stage is 2.5 times more sensitive to dicamba than the vegetative stages.

Environment by Rate.

Environment also impacts soybean's response to sub-lethal dicamba rates. ANOVA revealed an environment by rate interaction for yield reduction. Comparing among the three locations, the greatest yield reduction was observed at Kinston where yield reductions ranged from 10 to 80% as rate increased from 1.1 to 70 g ha^{-1} (Figure 10). At Lewiston, yield reductions ranged from 8 to 60%. Least yield reduction was observed at Rocky Mount, ranging from 1 to 62%. The lower yield reduction at Rocky Mount can be attributed to a planting date which was three weeks earlier than Kinston and Lewiston (Table #). When dicamba was applied at this location, soybean may have had more time to recover before maturity. When dicamba was applied at 8.8 g ha^{-1} (about 1/66th of the labeled rate for

dicamba-tolerant soybean), yield reduction was 20% or greater for all environments. Robinson et al. (2013) reported a yield loss of 12% when 5.6 g ha^{-1} of dicamba was applied to V7 soybeans.

Environment, Variety, and Application Timing.

ANOVA revealed a three-way interaction of environment, variety, and application timing, pooled over dicamba rates, for injury 14 and 28 DAT, height reduction 14 and 28 DAT, and yield reduction. When evaluated at 14 and 28 DAT, soybean injury was significantly greater at R2 applications, regardless of variety, compared to V4 applications for Rocky Mount (Table 4). Conversely, at Kinston, significantly greater injury 14 and 28 DAT was recorded when dicamba was applied at the V4 growth stage. At Lewiston, little difference due to application timing was observed 14 DAT while greater injury for the R2 application was observed for all varieties 28 DAT. When dicamba was applied V4, all varieties demonstrated the greatest injury 14 and 28 DAT at the Kinston location . For the R2 application, all varieties showed the greatest injury at Rocky Mount.

Among varieties, at Rocky Mount, SS 6810NR2 demonstrated significantly greater injury 14 DAT than CZ 6316LL for the V4 application (Table 4). For the R2 timing at the same location, injury for SS 6810NR2 was significantly greater than all varieties except SH 6515LL. There was no variety response for the V4 application at Kinston and the R2 application at Lewiston. For the R2 application at Kinston, the lowest injury was observed for SH 6515LL. At Lewiston, the greatest injury 14 DAT was observed for SS 6810NR2 when dicamba was applied V4. SS 6810NR2 generally exhibited the greatest visual injury 28 DAT with the exception of Kinston where no significant varietal response was observed.

At 28 DAT, height reduction followed the trend of Rocky Mount > Kinston > Lewiston. Analyzing among varieties grown in Lewiston, height reductions 28 DAT were not significantly different between growth stages at application, however significant varietal differences were observed (Table 4). Soybean grown at Lewiston showed significantly greater height reduction 28 DAT for DG S69RY34 when dicamba was applied V4 (Table 4). When dicamba was applied R2, SS 6810NR2 demonstrated the greatest height reduction compared to all other varieties. Comparing between application timings at Lewiston, height reduction for SS 6810NR2 was greater for the R2 timing compared to V4 while greater height reduction for the V4 application was observed for DG S69RY34.

In Rocky Mount, differences in injury were similar to those observed at 14 DAT with 3 of the 5 varieties showing significant differences between application timings. When dicamba was applied to DG S64LS95, DG S69RY34 and SH 6515LL, height reduction 28 DAT was greater for the V4 application and growth stage at time of application did not influence height reductions for SS 6810NR2 and DG S64LS95 (Table 4). At Rocky Mount, height reduction 28 DAT for CZ 6316LL was significantly greater than DG S64LS95 when dicamba was applied V4. For the R2 application height reductions were greatest for SS 6810NR2.

In Kinston, V4 height reductions were significantly greater than the R2 timing for all varieties (Table 4). Among varieties at Kinston, height reductions 28 DAT for SH 6515LL were significantly greater than CZ 6316LL and DG S69RY34 when dicamba was applied V4. For the R2 application, height reduction 28 DAT was greatest for SS 6810NR2.

Data analysis also revealed an environment, variety, and timing interaction for yield reduction. For trials conducted in Lewiston and Rocky Mount, all varieties, with the exception of CZ 6316LL at Lewiston, demonstrated greater sensitivity to dicamba when applications were made during the R2 growth stage (Table 4). At Lewiston, application timing did not influence yield reduction for CZ 6316LL. Interestingly, for the Kinston location, yield reduction for CZ 6316LL, DG S64LS95, and DG S69RY34 was generally greater for the R2 application but the differences due to application timing were not statistically significant. At the same location, yield reduction for SS 6810NR2 was significantly greater for the V4 application.

Examining among varieties at Lewiston, the greatest yield reduction for the V4 application was observed for SH 6515LL and DG S64LS95 while SS 6810NR2 had the lowest yield reduction (Table 4). When dicamba was applied R2 at the same location, DG S64LS95 and SS 6810NR2 demonstrated the greatest yield reduction. At the Rocky Mount location, yield reduction for all varieties was statistically similar for the V4 application. At the same location, when applications were made to the R2 stage, the greatest yield reduction was recorded for SS 6810NR2. At the Kinston location, when dicamba was applied V4, the greatest yield reduction was observed for SH 6515LL and SS 6810NR2 but no significant varietal response was noted for the R2 application.

Factors such as environmental conditions, soybean variety, and the growth stage at exposure all play a role in how soybeans will respond to dicamba drift. Although there was a great amount of variability among environments, SS 6810NR2 was consistently one of the most affected varieties when dicamba was applied R2. Previous research has concluded that

soybeans are more sensitive to dicamba at the reproductive growth stage than the vegetative stage (Wax et al. 1969, Griffin et al. 2013), and results from Lewiston and Rocky Mount generally agree with this conclusion. The majority of varieties, excluding CZ 6316LL, were more sensitive to dicamba applied R2.

The current research presents evidence that cultivar may influence soybean's response to dicamba drift but is ultimately dictated by factors within environments (i.e. growing conditions before during and after exposure or possibly planting date). Al-Khatib and Peterson (1999) reported increased soybean sensitivity when temperatures were high at time of exposure to dicamba. Factors such as humidity, time of day, rainfall, and ambient and soil temperature all have been analyzed but revealed no patterns that can explain the variability of the results. In some cases, specifically the R2 application at Kinston and V4 application at Rocky Mount, a varietal response was completely absent when dicamba was applied in certain environments and growth stages. More research should be done in controlled settings to understand what environmental factors impact dicamba activity when soybeans are exposed to sub-lethal doses of dicamba.

This research as well as previous research reaches contradicting conclusions. Although visual injury and height reductions have been considered to be moderate indicators of yield loss (Johnson, 2012; Weidenhamer, 1989), height reductions did not always translate to significant yield loss for V4 applications. When 8.8 g ha^{-1} of dicamba was applied to V4 soybeans in Rocky Mount, a height reduction of 30% was observed 28 DAT, however by the end of the year; yield reduction was only 4%. Also, when dicamba was applied when soybeans reached R2, significant yield reductions were recorded while no reductions in plant

height were observed. At Lewiston, no height reduction was recorded when 1.1 g ha^{-1} was applied R2 but yield reduction was 19%. Similarly, Auch and Arnold (1978) concluded that one could not always expect yield reduction when plant height is reduced from dicamba injury. The inconsistency in soybean response to simulated dicamba drift indicates strong environmental impacts, which make it difficult to predict yield loss. One conclusion that previous and current research can agree on is that low doses of dicamba have the potential to severely injure non-tolerant soybeans which can translate into significant yield losses. The varieties examined in this study cannot be used to minimize yield loss caused by off-target dicamba applications.

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Table 1. Planting date, harvest date and soil characteristics at experiment sites.

Location	Planting Date	Harvest Date	Year	Soil series	Textural classification	Soil subgroup	Soil pH[a]	Humic matter[a]
								%
Kinston	July16	January 3	2015	Portsmouth	Loam	Typic Umbraquults	6.2	2.37
Lewiston	July 13	January 5	2015	Rains	Sandy loam	Typic Paleaquults	5.9	0.92
Rocky Mount	June 22	November 15	2016	Rains	Sandy loam	Typic Paleaquults	5.8	0.60

[a] Soil pH and humic matter determined by the North Carolina Department of Agriculture and Consumer Services, Agronomic Division. Humic matter was determined photometrically according to Mehlich (1984).

Table 2. Recorded weather conditions during dicamba applications.

Location	Year	Application timing[a]	Application date	Relative humidity	Temperature	Soil Temperature
				%	°C	°C
Rocky Mount	2016	V4	July 19	85	36	30
		R2	August 10	82	33	28
Kinston	2015	V4	August 10	86	27	29
		R2	September 1	76	28	27
Lewiston	2015	V4	August 6	70	32	33
		R2	August 27	72	25	26

[a]Abbreviations: R2, reproductive stage; V4, vegetative stage.

Table 3. Pearson correlations among visual injury, height, and yield reduction of soybean.

Data are pooled over years, soils, varieties, and timings.

Variable	P>F	Correlation Coefficient
Injury 7 DAT[a] vs. Yield	<.0001	0.57
Injury 14 DAT vs. Yield	<.0001	0.59
Injury 28 DAT vs. Yield	<.0001	0.61
Height reduction 14 DAT vs. Yield	<.0001	0.41
Height reduction 28 DAT vs. Yield	<.0001	0.42

[a]Abbreviations: DAT, days after treatment.

Table 4. Soybean variety response to dicamba applied at V4 and R2 growth stages for injury, height reduction, and yield reduction, averaged over dicamba rates[abc].

Timing	Variety	Env[b]	Injury		Height[c]		Yield[c]
			14 DAT	28 DAT	14 DAT	28 DAT	
			--%--				
V4	CZ 6316LL	Kinston	44 bc	39 d	30 bc	31 bcd	35 e-i
		Lewiston	35 e-h	29 mno	14 ij	15 hij	29 g-k
		Rocky Mt	34 f-i	31 j-m	35 ab	35 ab	17 klm
	DG S64LS95	Kinston	44 bc	38 de	35 ab	36 ab	43 b-g
		Lewiston	32 jk	25 p	13 ij	14 ij	32 f-j
		Rocky Mt	37 ef	34 hij	28 b-e	27 c-f	20 j-m
	DG S69RY34	Kinston	42 cd	37 d-h	29 b-e	30 b-e	31 g-k
		Lewiston	34 f-i	29 mno	22 d-g	23 efg	19 klm
		Rocky Mt	35 e-h	32 jkl	29 b-e	31 bcd	10 lm
	SH 6515LL	Kinston	42 cd	38 def	40 a	41 a	51 a-d

Table 4 (Continued)

		Lewiston	33	hij	27	op	10	jkl	11	jkl	32	f-j
		Rocky Mt	35	e-h	32	jkl	29	bcd	30	b-e	11	lm
	SS 6810NR2	Kinston	45	bc	38	def	33	ab	34	abc	55	abc
		Lewiston	41	d	35	f-i	19	f-i	20	f-i	7	m
		Rocky Mt	38	e	35	f-i	30	bc	34	abc	23	i-l
R2	CZ 6316LL	Kinston	35	e-h	33	ijk	4	klm	6	klm	49	a-e
		Lewiston	37	ef	37	d-g	17	g-j	18	g-j	30	g-k
		Rocky Mt	43	bcd	45	c	22	d-g	25	d-g	42	b-h
	DG S64LS95	Kinston	32	jk	35	f-i	4	klm	5	klm	47	a-f
		Lewiston	35	e-h	36	e-i	13	ij	14	ij	57	ab
		Rocky Mt	44	bc	47	bc	25	c-f	26	def	40	c-h
	DG S69RY34	Kinston	35	e-h	35	f-i	11	jk	12	jk	45	a-g
		Lewiston	35	e-h	35	f-i	0	m	1	m	41	c-h
		Rocky Mt	44	bc	45	c	21	f-h	23	efg	27	h-k

Table 4 (Continued)

SH 6515LL	Kinston	30	k	31	j-m	11	jk	12	jk	43	b-g
	Lewiston	34	f-i	34	ghi	3	lm	4	lm	49	a-e
	Rocky Mt	46	ab	49	ab	16	g-j	22	fgh	35	e-i
SS 6810NR2	Kinston	36	efg	36	e-i	13	ij	14	ij	36	d-i
	Lewiston	37	ef	39	d	29	bcd	30	b-e	60	a
	Rocky Mt	48	a	51	a	31	bc	35	abc	55	abc

[a] Data averaged over dicamba rates. Means within a column followed by the same letter are not significantly different at $P = 0.05$ according to Fisher's Protected LSD.

[b] Abbreviations DAT, days after treatment; Env, environment.

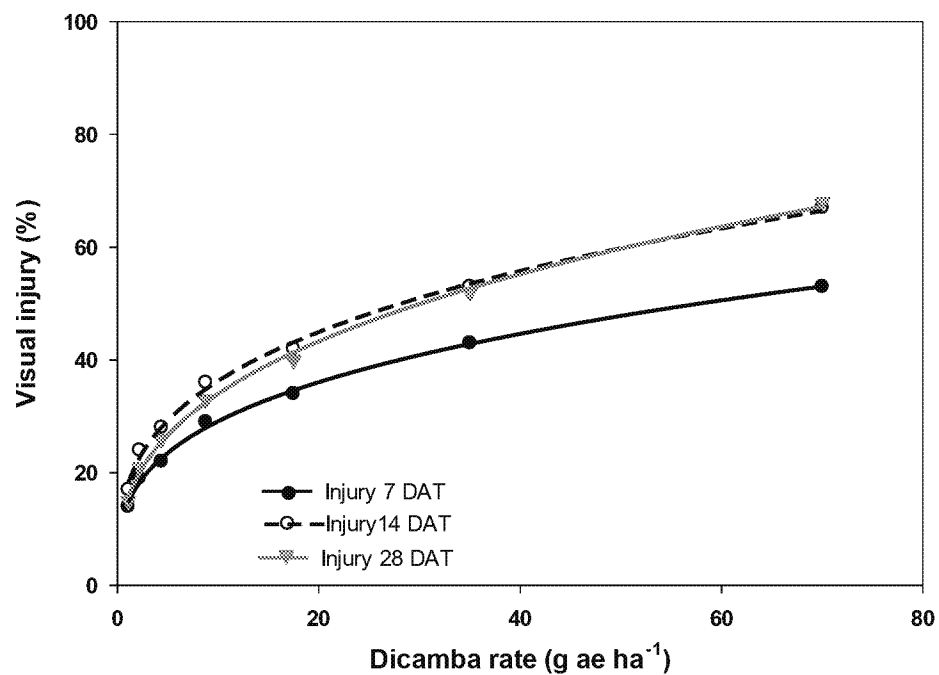[c] Expressed as a percentage of the non-treated control.

Figure 1. Soybean injury as affected by dicamba rate (1.1 to 70 g ha^{-1}) 7, 14, and 28 days after treatment (DAT). Predicted response 7 DAT can be described as Y= 14.3x$^{0.31}$, r^2=0.99. Predicted response 14 DAT can be described as Y=17.7x$^{0.31}$, r^2=0.99. Predicted response 28 DAT can be described as Y=15x$^{0.35}$, r^2=0.99.
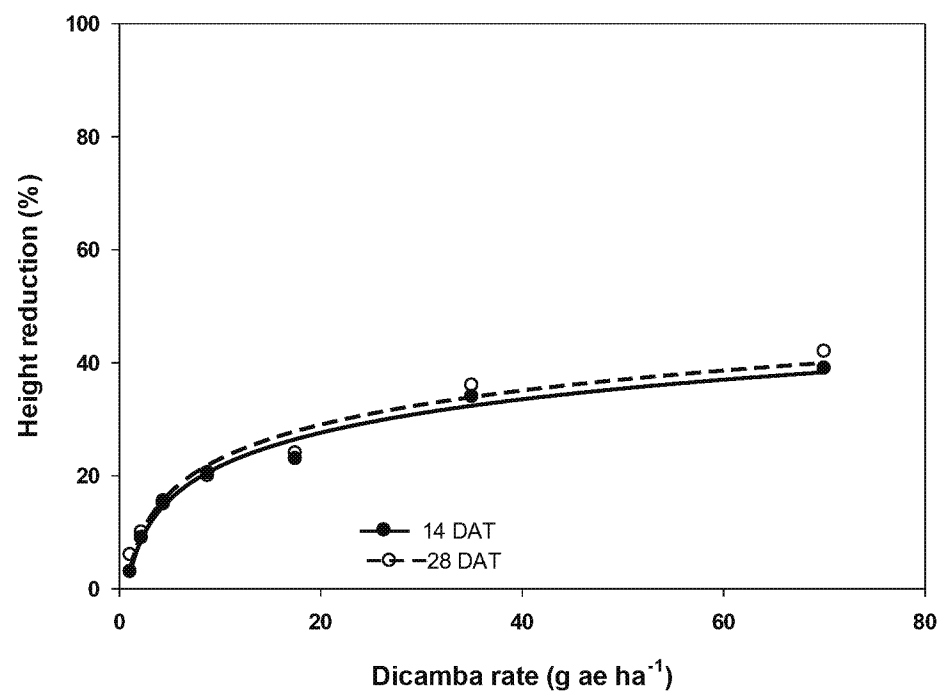
Figure 2. Soybean height reduction as affected by dicamba rate (1.1-70 g ae ha^{-1}) 14 and 28 days after treatment (DAT). Predicted response 14 DAT can be described as Y=2.0+8.5*ln (abs(x)), r^2=0.98. Predicted response 28 DAT can be described as Y=2.9+8.7*ln (abs(x)), r^2=0.96.
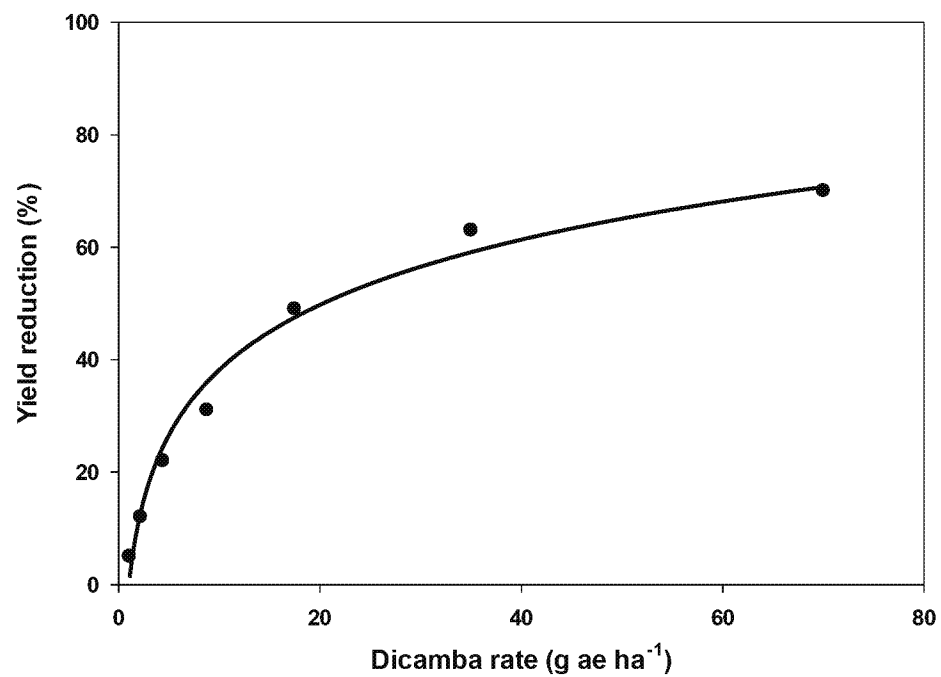
Figure 3. Soybean yield reduction as affected by dicamba rate (1.1-70 g ae ha^{-1}). Predicted yield response can be described as Y=-0.32+16.7*ln (abs(x)), r^2=0.98.

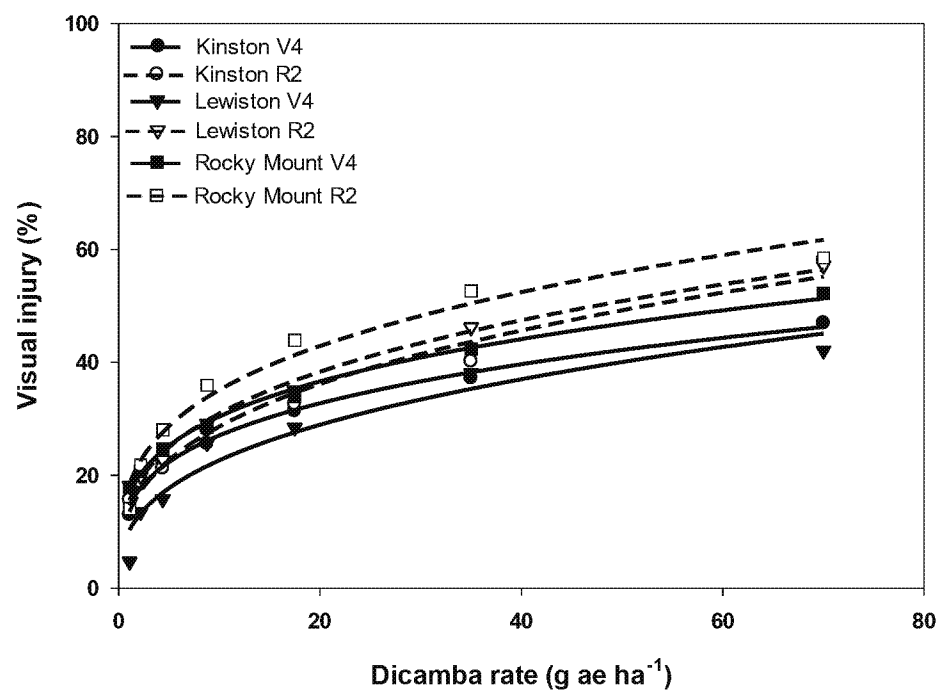Figure 4. Soybean injury as affected by dicamba rate, application timing and

environment 7 days after treatment (DAT). Predicted injury 7 DAT can be described

as: Kinston V4: $Y= 14.3x^{0.27}$, $r^2=0.98$. Kinston R2: $Y=13x^{0.33}$, $r^2=0.97$. Lewiston V4:

$Y=10x^{0.35}$, $r^2=0.93$. Lewiston R2: $Y=15x^{0.31}$, $r^2=0.99$. Rocky Mount V4: $Y=16x^{0.27}$,

$r^2=0.99$. Rocky Mount R2: $Y=18x^{0.29}$, $r^2=0.96$.

Figure 5. Soybean injury as affected by dicamba rate, application timing and environment 14 DAT. Predicted injury 14 DAT can be described as: Kinston V4: $Y= 27x^{0.20}$, $r^2=0.98$. Kinston R2: $Y=11.5x^{0.41}$, $r^2=0.97$. Lewiston V4: $Y=16x^{0.32}$, $r^2=0.90$. Lewiston R2: $Y=14x^{0.37}$, $r^2=0.97$. Rocky Mount V4: $Y=18.5x^{0.27}$, $r^2=0.99$. Rocky Mount R2: $Y=21x^{0.31}$, $r^2=0.98$.
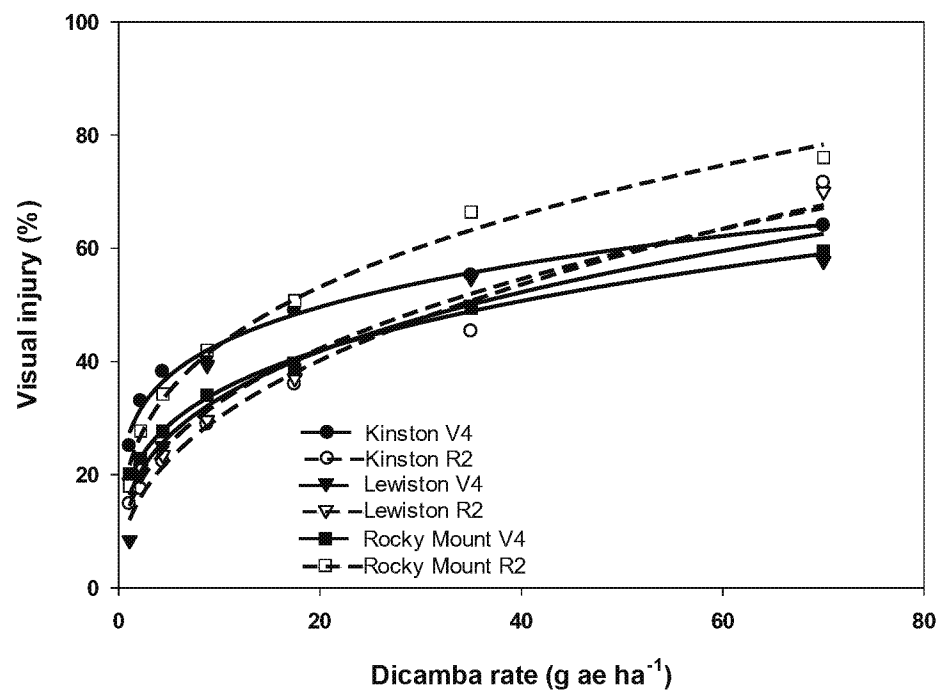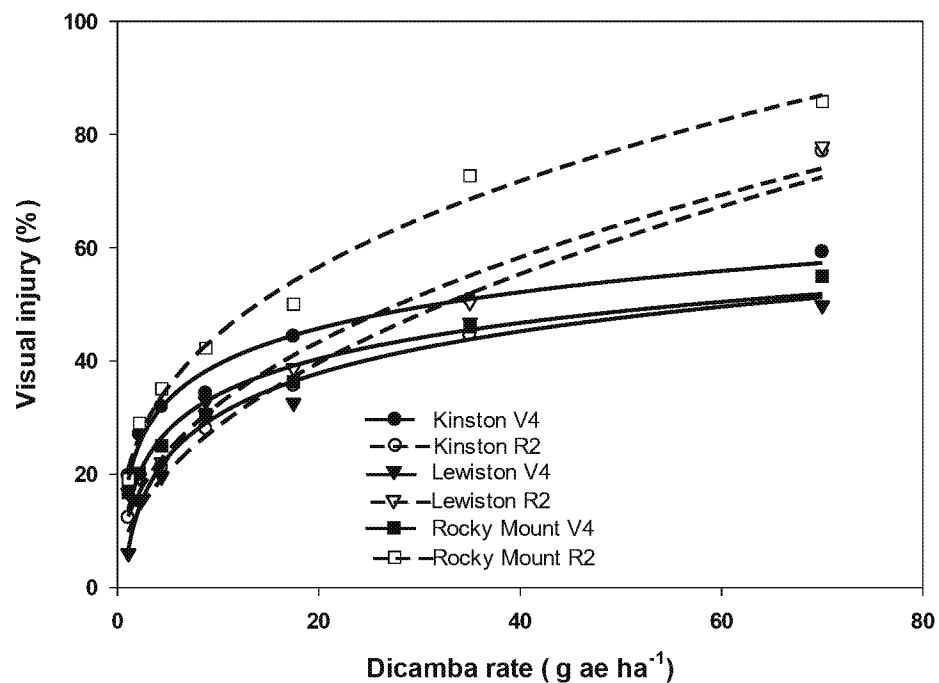
Figure 6. Soybean injury as affected by dicamba rate, application timing and

environment 28 DAT. Predicted injury 28 DAT can be described as: Kinston V4:

Y= 18+9*ln(abs(x)), r^2=0.98. Kinston R2: Y=5.8+10.7*ln (abs(x)), r^2=0.96.

Lewiston V4: Y=13+9.2*ln (abs(x)), r^2=0.96. Lewiston R2: Y=20x$^{0.34}$, r^2=0.98.

Rocky Mount V4: Y=9x$^{0.48}$, r^2=0.96. Rocky Mount R2: Y=12x$^{0.43}$, r^2=0.97.
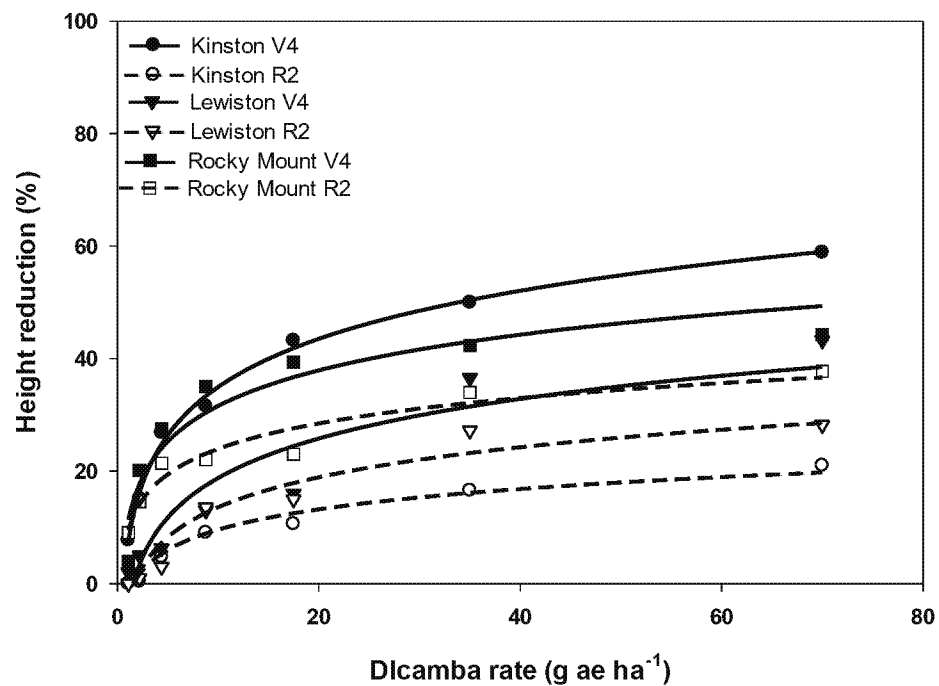
Figure 7. Soybean height reduction as affected by dicamba rate, application timing and

environment 14 DAT. Predicted height reduction 14 DAT can be described as: Kinston V4:

Y= 6.5+12*ln(abs(x)), r^2=0.99. Kinston R2: Y=-2.4+5*ln (abs(x)), r^2=0.96. Lewiston V4:

Y=-4.6+10*ln (abs(x)), r^2=0.85. Lewiston R2: Y=-4+7.7*ln (abs(x)), r^2=0.92. Rocky Mount

V4: Y=10+9*ln (abs(x)), r^2=0.88. Rocky Mount R2: Y=9+6.5*ln (abs(x)), r^2=0.93.
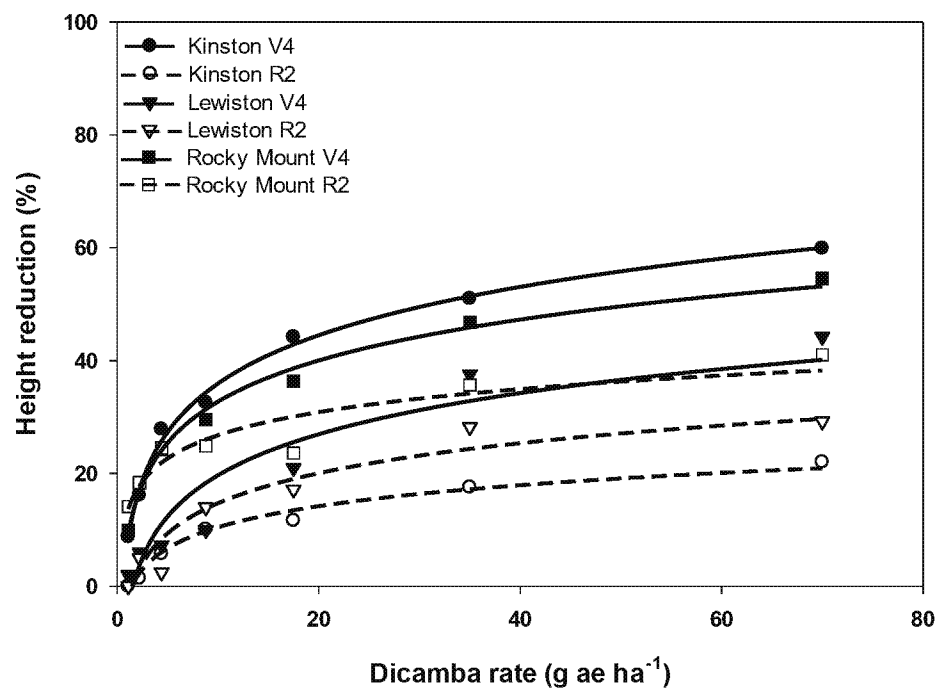
Figure 8. Soybean height reduction as affected by dicamba rate, application timing and environment 14 DAT. Predicted height reduction 14 DAT can be described as: Kinston V4: Y= 7.5+12*ln (abs(x)), r^2=0.99. Kinston R2: Y=-1.9+5*ln (abs(x)), r^2=0.98. Lewiston V4: Y=-4.5+10.5*ln (abs(x)), r^2=0.87. Lewiston R2: Y=-3+7.7*ln (abs(x)), r^2=0.91. Rocky Mount V4: Y=13+6*ln (abs(x)), r^2=0.86. Rocky Mount R2: Y=8.5+10.5*ln (abs(x)), r^2=0.99.
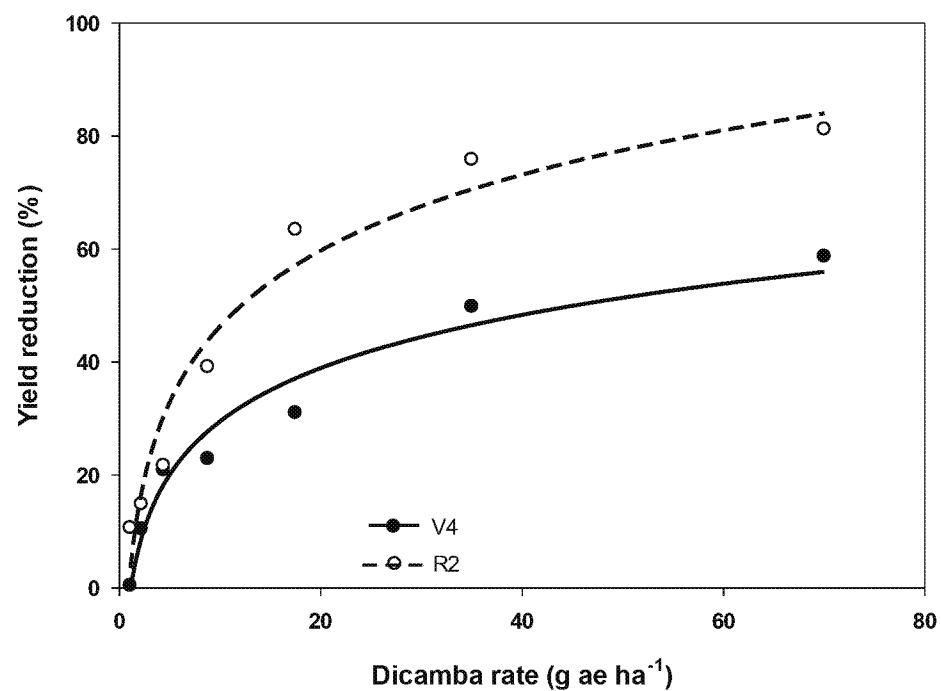
Figure 9. Yield reduction as affected by dicamba rate (1.1-70 g ha^{-1}) and application timing. Predicted yield reduction for V4 application can be described as: Y= -1.8+13.6*ln(abs(x)) , r^2=0.94. Predicted yield reduction for the R2 application can be described as: Y=1.7+19.3* ln(abs(x)), r^2= 0.96.
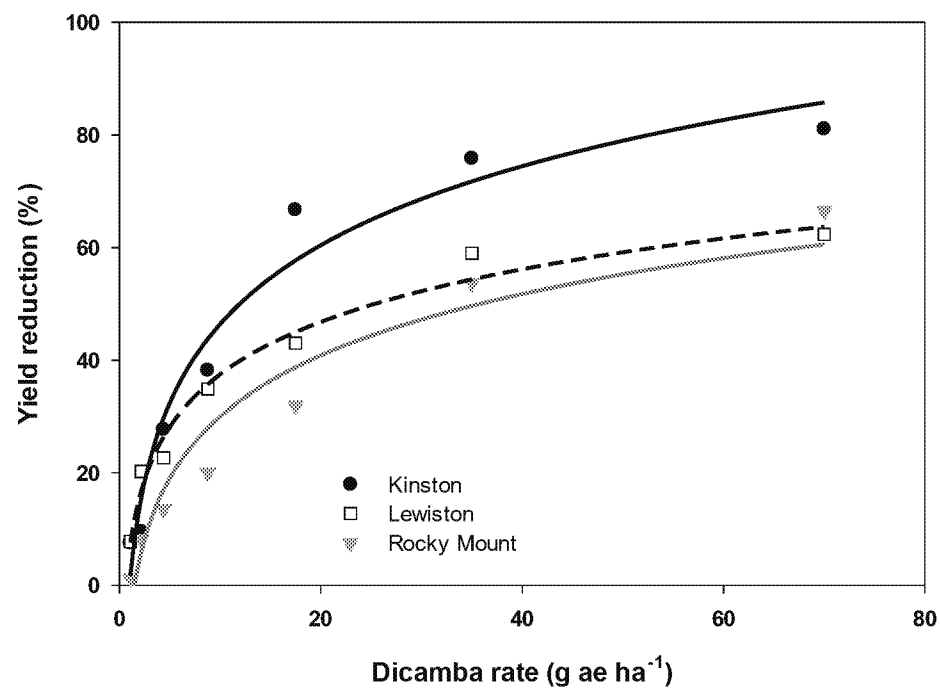
Figure 10. Yield reduction as affected by dicamba rate (1.1-70 g ha^{-1}) and environment.

Predicted yield reduction for Kinston can be described as: Y= -0.2+20*ln (abs(x)), r^2=0.95.

Predicted yield reduction for Lewiston can be described as: Y=6.4+13.5*ln (abs(x)), r^2=0.97.

Predicted yield reduction for Rocky Mount can be described as Y=-6.3+15.7*ln (abs(x)),

r^2=0.93

APPENDICES

CHAPTER II ANOVA TABLES

Table 1. Analysis of variance for soybean injury.[a]

Source	df	7 DAT		14 DAT		28 DAT	
		Mean square	p-value	Mean square	p-value	Mean square	p-value
Soil	1	60	0.1177	772	<0.0001	1927	<0.0001
Year	1	1542	<0.0001	87	0.0844	28	0.3336
Soil*Year	1	297	0.0005	56	0.1634	9	0.5789
Variety (Var)	3	185	<0.0001	469	<0.0001	1017	<0.0001
Soil*Var	3	110	0.0037	34	0.3172	31	0.3677
Year*Var	3	299	0.0001	412	<0.0001	755	<0.0001
Soil*Var*Year	3	76	0.0260	39	0.2535	85	0.0348
Rate	6	27804	<0.0001	43113	<0.0001	47888	<0.0001
Soil*Rate	6	149	<0.0001	403	<0.0001	994	<0.0001
Year*Rate	6	203	<0.0001	103	0.0019	170	<0.0001
Soil*Year*Rate	6	316	<0.0001	354	<0.0001	272	<0.0001
Var*Rate	18	27	0.3536	29	0.4772	27	0.5393
Soil*Var*Rate	18	20	0.6721	19	0.8542	30	0.4360
Year*Var*Rate	18	46	0.0147	42	0.1057	39	0.1613

Table 1 (Continued)

Soil*Year*Var*Rate	18	30	0.2318	20	0.8133	25	0.6286
Growth stage (GS)	1	4139	<0.0001	31355	<0.0001	9613	<0.0001
Soil*GS	1	4282	<0.0001	4462	<0.0001	6012	<0.0001
Year*GS	1	4153	<0.0001	9	0.5787	64	0.1399
Soil*Year*GS	1	158	0.0110	105	0.0577	90	0.0803
Var*GS	3	205	<0.0001	552	<0.0001	646	<0.0001
Soil*Var*GS	3	112	0.0033	367	<0.0001	366	<0.0001
Year*Var*GS	3	20	0.4819	77	0.0466	127	0.0050
Soil*Year*Variety*GS	3	27	0.3494	61	0.0971	141	0.0027
Rate*GS	6	257	<0.0001	230	<0.0001	28	0.4573
Soil*Rate*GS	6	257	<0.0001	568	<0.0001	850	<0.0001
Year*Rate*GS	6	76	0.0050	400	<0.0001	227	<0.0001
Soil*Year*Rate*GS	6	27	0.3512	13	0.8308	11	0.8893
Var*Rate*GS	18	35	0.1071	40	0.1344	35	0.2582
Soil*Var*Rate*GS	18	28	0.2905	51	0.0258	45	0.0792
Year*Var*Rate*GS	18	19	0.7098	40	0.1304	42	0.1053
Soil*Year*Var*Rate*GS	18	16	0.8373	46	0.0560	48	0.0477

[a] Abbreviations: df, degrees of freedom; DAT, days after treatment.

Table 2. Analysis of variance for soybean height reduction and yield reduction.[a]

| | | Height reduction | | | | Yield reduction | |
| | | 14 DAT | | 28 DAT | | | |
Source	df	Mean square	p-value	Mean square	p-value	Mean square	p-value
Soil	1	19	0.6852	0.65	0.9426	2665	0.0497
Year	1	1090	0.0020	49	0.5357	472	0.4085
Soil*Year	1	18	0.6890	1195	0.0022	18054	<0.0001
Variety (Var)	3	845	<0.0001	613	0.0024	2374	0.0165
Soil*Var	3	1083	<0.0001	191	0.2116	3074	0.0041
Year*Var	3	849	<0.0001	578	0.0036	679	0.3996
Soil*Var*Year	3	1646	<0.0001	648	0.0017	4636	0.0002
Rate	6	18332	<0.0001	26255	<0.0001	106317	<0.0001
Soil*Rate	6	41	0.9020	99	0.5862	283	0.8720
Year*Rate	6	459	0.0005	146	0.3306	1323	0.0753
Soil*Year*Rate	6	94	0.5490	187	0.1840	724	0.3914
Var*Rate	18	81	0.8026	73	0.9154	911	0.1672
Soil*Var*Rate	18	97	0.6352	136	0.3779	935	0.1468

Table 2 (Continued)

Year*Var*Rate	18	110	0.4977	101	0.7047	603	0.6108
Soil*Year*Var*Rate	18	40	0.9945	34	0.9991	859	0.2180
Growth stage (GS)	1	68081	<0.0001	89157	<0.0001	8641	0.0004
Soil*GS	1	823	0.0072	1867	0.0001	27196	<0.0001
Year*GS	1	3589	<0.0001	746	0.0154	1376	0.1582
Soil*Year*GS	1	224	0.1599	3451	<0.0001	1047	0.2182
Var*GS	3	2666	<0.0001	3725	<0.0001	1238	0.1468
Soil*Var*GS	3	481	0.0055	382	0.0294	263	0.7663
Year*Var*GS	3	913	<0.0001	2181	<0.0001	3930	0.0007
Soil*Year*Variety*GS	3	74	0.5836	611	0.0025	1482	0.0928
Rate*GS	6	201	0.1031	499	0.0007	894	0.2561
Soil*Rate*GS	6	144	0.2678	264	0.0528	789	0.3348
Year*Rate*GS	6	591	<0.0001	829	<0.0001	201	0.9407
Soil*Year*Rate*GS	6	140	0.2850	82	0.6925	1281	0.0857
Var*Rate*GS	18	57	0.9555	32	0.9994	787	0.3061
Soil*Var*Rate*GS	18	36	0.9971	99	0.7204	813	0.2722
Year*Var*Rate*GS	18	28	0.9995	42	0.9963	223	0.9967

Table 2 (Continued)

Soil*Year*Var*Rate*GS	18	58	0.9540	63	0.9605	561	0.6848

[a] Abbreviations: df, degrees of freedom; DAT, days after treatment.

CHAPTER III ANOVA TABLES

Table 1. Analysis of variance of percent injury 7 DAT[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	115.15	<.0001*
Rate	6	1020.03	<.0001*
Env*Rate	12	4.39	<.0001*
Variety (var)	4	10.14	<.0001*
Env*Var	8	2.63	0.0077*
Rate*Var	24	1.97	0.0039*
Env*Rate*Var	48	0.65	0.9673
Timing	1	266.33	<.0001*
Env*Timing	2	29.78	<.0001*
Rate*Timing	6	11.96	<.0001*
Env*Rate*Timing	12	7.77	<.0001*
Var*Timing	4	1.01	0.401
Env*Var*Timing	8	1.87	0.0614
Rate*Var*Timing	24	1.19	0.2436
Env*Rate*Var*Timing	48	0.92	0.6319

[a]Abbreviations: df, degrees of freedom; DAT, days after treatment.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.

Table 2. Analysis of variance of percent injury 14 DAT[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	70.46	<.0001*
Rate	6	1332.2	<.0001*
Env*Rate	12	4.84	<.0001*
Variety (Var)	4	15.8	<.0001*
Env*Var	8	3.63	0.0004*
Rate*Var	24	2.11	0.0016*
Env*Rate*Var	48	0.91	0.6532
Timing	1	0.02	0.8768
Env*Timing	2	233.1	<.0001*
Rate*Timing	6	36.04	<.0001*
Env*Rate*Timing	12	14.42	<.0001*
Var*Timing	4	0.69	0.5997
Env*Var*Timing	8	3.37	0.0009*
Rate*Var*Timing	24	1.3	0.1534
Env*Rate*Var*Timing	48	1.19	0.188

[a]Abbreviations: df, degrees of freedom; DAT, days after treatment.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.

Table 3. Analysis of variance of percent injury 28 DAT[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	149.64	<.0001*
Rate	6	1528.23	<.0001*
Env*Rate	12	5.58	<.0001*
Variety (Var)	4	18.47	<.0001*
Env*Var	8	3.92	0.0002*
Rate*Var	24	1.51	0.0573
Env*Rate*Var	48	1	0.4725
Timing	1	272.97	<.0001*
Env*Timing	2	262.04	<.0001*
Rate*Timing	6	92.93	<.0001*
Env*Rate*Timing	12	10.86	<.0001*
Var*Timing	4	0.12	0.9762
Env*Var*Timing	8	4.59	<.0001*
Rate*Var*Timing	24	1.42	0.0894
Env*Rate*Var*Timing	48	1.06	0.3736

[a]Abbreviations: df, degrees of freedom; DAT, days after treatment.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.

Table 4. Analysis of variance of percent height reduction 14 DAT[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	51.88	<.0001*
Rate	6	93.75	<.0001*
Env*Rate	12	2.51	0.0031*
Variety (Var)	4	7.78	<.0001*
Env*Var	8	4.52	<.0001*
Rate*Var	24	0.95	0.5279
Env*Rate*Var	48	0.64	0.9722
Timing	1	138.33	<.0001*
Env*Timing	2	42.42	<.0001*
Rate*Timing	6	4.46	0.0002*
Env*Rate*Timing	12	3.19	0.0002*
Var*Timing	4	5.34	0.0003*
Env*Var*Timing	8	4.46	<.0001*
Rate*Var*Timing	24	0.59	0.9405
Env*Rate*Var*Timing	48	1.06	0.3736

[a]Abbreviations: df, degrees of freedom; DAT, days after treatment.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.

Table 5. Analysis of variance of percent height reduction 28 DAT[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	60.76	<.0001*
Rate	6	98.12	<.0001*
Env*Rate	12	1.47	0.1319
Variety (Var)	4	8.78	<.0001*
Env*Var	8	4.26	<.0001*
Rate*Var	24	0.85	0.6784
Env*Rate*Var	48	0.57	0.9912
Timing	1	121.81	<.0001*
Env*Timing	2	46.02	<.0001*
Rate*Timing	6	6.61	<.0001*
Env*Rate*Timing	12	2.13	0.0135*
Var*Timing	4	4.69	0.001*
Env*Var*Timing	8	4.27	<.0001*
Rate*Var*Timing	24	0.96	0.5217
Env*Rate*Var*Timing	48	0.48	0.9989

[a]Abbreviations: df, degrees of freedom; DAT, days after treatment.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.

Table 6. Analysis of variance of percent yield reduction[a].

Source	df[a]	F-value	p-value
Environment (Env)	2	21.27	<.0001*
Rate	6	86.01	<.0001*
Env*Rate	12	2.32	0.0067*
Variety (Var)	4	4.21	0.0023*
Env*Var	8	1.84	0.0671
Rate*Var	24	0.91	0.5928
Env*Rate*Var	48	0.8	0.837
Timing	1	62.25	<.0001*
Env*Timing	2	14.2	<.0001*
Rate*Timing	6	4.59	0.0001*
Env*Rate*Timing	12	1.73	0.0572
Var*Timing	4	0.82	0.5158
Env*Var*Timing	8	4.24	<.0001*
Rate*Var*Timing	24	1.4	0.0985
Env*Rate*Var*Timing	48	1.05	0.392

[a]Abbreviations: df, degrees of freedom.
[b]An asterisk denotes significance at the $\alpha = 0.05$ level according to Fisher's Protected LSD.